UNIVERSITÉ DU QUÉBEC

DÉTERMINANTS DE L'EXPRESSION DU POLYMORPHISME ASSOCIÉ AUX RESSOURCES CHEZ L'OMBLE DE FONTAINE, *SALVELINUS FONTINALIS*, DANS LES LACS DU BOUCLIER LAURENTIEN

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Les chapitres II, III, et IV de cette thèse sont présentés sous forme d'articles scientifiques. Les données analysées dans ces trois chapitres proviennent des travaux de terrain réalisés durant les étés 2012, 2013 et 2014 pour lesquels j'ai activement participé. J'ai contribué à l'élaboration du plan d'échantillonnage en collaboration avec mon directeur de recherche (Pierre Magnan) ainsi qu'avec Marc Pépino (professionnel de recherche). La logistique ainsi que l'encadrement des assistants de recherche ont relevé de la responsabilité de Pierre Magnan, Marc Pépino et moi-même. Je confirme avoir rédigé toutes les premières versions des articles présentés et intégré les révisions et suggestions faites par mes co-auteurs suite à leur lecture des manuscrits. Ces trois articles scientifiques ont été ou seront soumis à des revues scientifiques avec comités de lecture. Le premier article (Chapitre II) a été soumis au périodique *Journal of Animal Ecology* et l'éditeur nous a invité à resoumettre une version révisée pour une nouvelle évaluation par les arbitres. Le deuxième article (Chapitre III) est actuellement sous presse dans le périodique *Oecologia* et devrait être publié au début de 2021. Enfin, le troisième article (Chapitre IV) est a été soumis au périodique *Oecologia* et est actuellement en révision.

RÉSUMÉ

Comprendre les mécanismes menant à la l'émergence de nouvelles espèces (c.-à-d., la spéciation) est au cœur de l'écologie évolutive et nous avons longtemps pensé que ce phénomène se produisait principalement en allopatrie (c.-à-d., suite à l'isolement géographique d'une population) sur une échelle temporelle très longue. Or, plusieurs études récentes suggèrent plutôt que ces processus évolutifs peuvent prendre place sur une courte période de temps et que le rôle de diversification de la spéciation sympatrique a largement été sous-estimé. Pour identifier et mieux comprendre les mécanismes responsables de la spéciation sympatrique, il est possible d'utiliser à notre avantage les populations présentant une spéciation incomplète. Le long de ce gradient de spéciation, nous retrouvons les populations exprimant un polymorphisme associé aux ressources. Ce phénomène se manifeste lorsque les individus d'une même population expriment des phénotypes distincts associés à une utilisation différentielle des ressources.

L'omble de fontaine, *Salvelinus fontinalis*, affiche un polymorphisme associé aux ressources dans les lacs du Bouclier laurentien, où un écotype littoral s'alimente de proies benthiques dans la zone littorale alors qu'un écotype pélagique s'alimente de zooplancton dans la colonne d'eau. Cependant, les connaissances acquises jusqu'à ce jour dans ce système l'ont été sur un nombre restreint de lacs et ont fait ressortir que l'expression du polymorphisme associé aux ressources est variable d'un lac à l'autre, en termes d'occurrence et de différences morphologiques entre les deux écotypes. L'objectif de ce projet a donc été de déterminer l'influence de la compétition intra- et interspécifique ainsi que des facteurs environnementaux sur l'expression du polymorphisme associé aux ressources chez l'omble de fontaine à plus large échelle.

Pour ce faire, nous avons échantillonné près d'une trentaine de populations d'omble de fontaine lacustres du Bouclier laurentien n'ayant jamais été ensemencées. Les lacs sélectionnés présentaient différents niveaux de compétition interspécifique, c'est-à-dire des lacs où l'omble de fontaine était la seule espèce de poisson présente, des lacs où il était en sympatrie avec du mulet à cornes, *Semotilus atromaculatus*, et des lacs où l'on retrouvait également du meunier noir, *Catostomus commersonii*). Les poissons ont été capturés à l'aide de filets-trappes installés dans les zones littorale et pélagique pour une période de cinq jours consécutifs de mai à août pour les années 2012 à 2014.

Dans un premier temps, nous avons évalué l'impact de la compétition interspécifique et de plusieurs facteurs environnementaux sur l'abondance des ombles de fontaine dans chacun des habitats (littoral et pélagique) ainsi que leurs effets sur la sélection d'habitats littoral et pélagique à l'aide d'une approche basée sur la théorie des isodars. Nos résultats montrent un effet clair de la compétition interspécifique par le meunier noir sur l'abondance des ombles de fontaine, et ce, autant en zone littorale que pélagique. Toutefois, aucun effet sur l'abondance des ombles de fontaine n'a été détecté en réponse à la présence de mulet à cornes. De plus, nos résultats suggèrent un effet de densité-dépendance dans la sélection d'habitat entre les habitats littorale et pélagique où les

ombles de fontaine préfèrent l'habitat littoral lorsque la température y est optimale. Ces analyses montrent également que l'augmentation de la température de l'épilimnion affecte de manière drastique cette relation de densité-dépendance, les ombles évitant l'habitat littoral lorsque l'épilimnion dépasse 22.2°C. Le réchauffement des lacs anticipé en raison des changements climatiques pourrait donc réduire l'accessibilité aux ressources littorales et ainsi perturber l'expression du polymorphisme chez l'omble de fontaine.

Dans un second temps, nous avons évalué comment ces facteurs influencent la spécialisation dans l'utilisation des ressources alimentaires des écotypes littoral et pélagique. Pour ce faire, nous avons sélectionné cinq indicateurs de l'utilisation des ressources présentant différents temps d'intégration (c.-à-d., contenus stomacaux, signature isotopique du carbone dans le foie, concentration en caroténoïdes dans les muscles, longueur des caeca pyloriques et longueur des branchicténies). Nos résultats montrent une spécialisation alimentaire claire des deux écotypes pour tous les indicateurs, à l'exception des branchicténies, suggérant que le polymorphisme associé aux ressources est un phénomène stable chez l'omble de fontaine retrouvé dans ces écosystèmes. De plus, tous les indicateurs, sauf la signature isotopique du foie, indiquent que la compétition interspécifique pousse les ombles à incorporer davantage de ressources pélagiques dans leur alimentation. Contrairement à notre hypothèse de départ, la pression de compétition interspécifique n'affecte pas l'amplitude des différences dans l'utilisation des ressources entre les deux écotypes. Nous croyons que les différences dans le mode d'alimentation entre des compétiteurs distants au niveau phylogénétique pourraient limiter l'exclusion compétitive complète d'une espèce et expliquer l'absence de réponse en lien avec le relâchement écologique.

Dans un troisième temps, nous avons vérifié si les écotypes littoral et pélagique présentent une évolution parallèle (c.-à-d., si les mêmes caractéristiques morphologiques distinguent les deux écotypes dans l'ensemble des lacs étudiés) et comment la compétition interspécifique en zone littorale affecte ces différences, le cas échéant. Pour ce faire, nous avons utilisé une combinaison de traits morphologiques ainsi que la forme du corps afin de caractériser les différences morphologiques entre les écotypes littoral et pélagique de populations, le long du gradient de compétition interspécifique présenté ci-haut. Nos résultats suggèrent que l'amplitude des différences morphologiques entre les écotypes est faible dans les populations lacustres d'omble de fontaine du Bouclier laurentien. Cependant, nous avons tout de même observé un certain niveau d'évolution parallèle caractérisée par des individus en moyenne plus trapu, avec de plus longues nageoires et de plus petits yeux pour l'écotype littoral comparativement à l'écotype pélagique. De plus, nos résultats montrent que la compétition interspécifique ne semble pas affecter l'amplitude ou la nature des différences morphologiques entre les écotypes, mais plutôt la biomasse d'omble de fontaine (dans les lacs où le meunier noir est présent) et leur condition physique (omble de fontaine plus maigre dans les lacs avec seulement le mulet à cornes comme compétiteur).

En conclusion, dans leur ensemble, nos résultats indiquent que la compétition intraspécifique est forte dans ce système et que contrairement à la théorie, la compétition interspécifique ne semble pas réduire la spécialisation alimentaire et morphologique des deux écotypes. De plus, nos résultats montrent que le meunier noir a un effet important

sur l'abondance relative de l'omble de fontaine alors que le mulet à cornes semble davantage affecter leur condition physique. Finalement, bien que nous ayons observé une spécialisation au niveau des ressources utilisées par les écotypes littoral et pélagique, la forte variation dans les traits impliqués dans la discrimination des deux écotypes et le faible niveau d'évolution parallèle suggèrent que les conditions physiques et écologiques sont variables d'un lac à l'autre et façonneraient les caractéristiques morphologiques à une échelle plus locale qu'anticipée. Ainsi, la présence de spécialisations alimentaires claires entre les écotypes et l'absence de différences marquées entre les caractéristiques morphologiques des deux écotypes suggèrent que l'omble de fontaine présente un stade de diversification très précoce. Par l'étude simultanée de l'impact de la compétition intra- et interspécifique ainsi que de plusieurs facteurs environnementaux sur l'expression du polymorphisme associé aux ressources, et ce, à une échelle permettant de généraliser les connaissances actuelles, nous croyons que ces travaux auront une incidence importance en écologie fonctionnelle et évolutive.

Mots-clés : Spéciation, plasticité phénotypique, distribution idéale libre, isodar, thermorégulation comportementale, spécialisation individuelle, morphologie, sélection d'habitat, salmonidé.

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CHAPITRE I

INTRODUCTION

1.1 Les différences individuelles comme fondation de l'écologie évolutive

Il est reconnu depuis longtemps que les espèces animales diffèrent dans l'utilisation qu'elles font de leurs habitats (« niche écologique de Hutchinson »; Hutchinson 1957). Cependant, la plupart des études négligent la possibilité d'une utilisation différente des ressources entre les individus d'une même espèce (Bolnick et al. 2003). Bien que parfois cette simplification est nécessaire lors de l'analyse de phénomènes complexes, elle peut également avoir comme conséquence de sous-représenter l'importance des différences individuelles dans les processus écologiques et évolutifs (Bolnick et al. 2003). La présence de différences entre les individus d'une même espèce n'est pourtant pas une découverte nouvelle (Darwin 1859; Van Valen 1965) et plusieurs études récentes suggèrent que les différences individuelles sont à la base de nombreux processus écologiques et évolutifs (Bolnick et al. 2010; Araújo et al. 2011; Svanbäck et al. 2015). Bien que dans plusieurs cas, ces différences individuelles sont associées à l'ontogénie (p. ex., Nakazawa 2015) ou à un dimorphisme sexuel (p. ex., Shine 1989), nous nous intéressons ici plus particulièrement aux différences individuelles indépendantes de ces phénomènes, que nous appellerons ci-après « spécialisation individuelle ». Comme ces spécialisations individuelles dans l'utilisation des ressources sont souvent associées à des variations comportementales, morphologiques et/ou physiologiques (Bolnick et al. 2003; Dall et al. 2012), ce phénomène pourrait avoir d'importantes implications évolutives en favorisant la variation phénotypique indispensable aux processus de sélection naturelle. De telles spécialisations pourraient donc constituer une pierre angulaire de la biodiversité en agissant comme base pour les processus de spéciation sympatrique (Bolnick et al. 2003).

Il est possible de diviser l'étude des spécialisations individuelles dans l'utilisation des ressources en deux grands cadres théoriques complémentaires, qui se distinguent

principalement par la nature des ressources/habitats qui caractérisent la spécialisation des individus (Smith et Skùlason 1996; Bolnick et al. 2003). Lorsque les ressources ou les habitats peuvent être caractérisés le long d'un gradient continu (p. ex., longueur des proies), Roughgarden (1974) a proposé une première mesure quantitative de spécialisation alimentaire WIC/TNW (angl., within individual component / total niche width) qui met en relation la variance moyenne des ressources utilisées par chaque individu (que l'on peut qualifier de composante intra-individuelle ou WIC) avec la variance totale des ressources utilisées par la population (que l'on peut qualifier comme étant l'étendue totale de la niche d'une population ou TNW). L'étendue totale de la niche (TNW) peut à son tour être divisée en deux composantes, soit la WIC et la composante inter-individuelle (BIC) qui se caractérise comme la variation dans l'utilisation des ressources entre les individus. Ainsi, plus la composante inter-individuelle (BIC) est élevée en comparaison à la composante intra-individuelle, plus le ratio (c.-à-d., WIC/TNW) sera petit et donc plus la spécialisation individuelle sera considérée élevée. Cette approche a également été adaptée à l'utilisation de données catégoriques de proies en utilisant l'indice de diversité de Shannon en remplacement de la variance (Roughgarden 1979). Bien que ce cadre conceptuel offre plusieurs avantages, notamment sa simplicité, il est cependant difficile à appliquer lorsque les ressources et habitats utilisés par une population sont discrets (c.-à-d., distincts et fonctionnellement différents). De plus, comme suggérées par Smith et Skúlason (1996), de telles ressources (discrètes) entraînent souvent des spécialisations individuelles menant à la formation d'écotypes distincts se spécialisant sur certaines ressources. Telle que soulignée par Bolnick et al (2003), la distinction entre la spécialisation individuelle et le polymorphisme associé aux ressources est principalement sémantique. En effet, bien que ces deux termes soient utilisés pour décrire des phénomènes où les individus d'une même population utilisent une fraction des ressources disponibles, le polymorphisme associé aux ressources se caractérise également par la présence d'écotypes aux phénotypes généralement distincts (Smith et Skúlason 1996). Ainsi, bien que ces deux phénomènes présentent de grandes similarités au niveau mécanistique et conceptuel, ils constituent les extrêmes d'un même gradient allant de variations continues (spécialisation individuelle) à discrètes (polymorphisme associé aux ressources) (Bolnick et al. 2003). Dans le cadre de cette thèse, nous nous intéresserons

davantage au polymorphisme associé aux ressources et bien que ces phénomènes comportent des composantes génotypiques et phénotypiques, nous avons également mis l'accent sur l'analyse du phénotype en raison de l'effet direct des forces de sélection naturelle sur ce dernier (Hendry 2016). En effet, ce n'est qu'à travers l'expression phénotypique que les forces de sélection peuvent avoir un impact sur le génotype (Hendry 2016; Dall et al. 2012).

1.2 Polymorphisme associé aux ressources

Dans le cadre de cette thèse, nous définissons le polymorphisme associé aux ressources comme l'occurrence de phénotypes discrets (écotypes) au sein d'une même population qui présentent une utilisation différentielle des ressources. Les écotypes peuvent présenter des différences morphologiques, comportementales, pigmentaires ou encore au niveau des traits d'histoire de vie (Smith et Skúlason 1996). Le polymorphisme associé aux ressources serait fréquent chez les vertébrés (Smith et Skúlason 1996) et constituerait une étape importante dans le processus de spéciation sympatrique (Wilson 1998, Barluenga et al. 2006, Pfennig et al. 2010). En effet, plusieurs études qui se sont intéressées aux mécanismes responsables du polymorphisme associé aux ressources ont relevé la présence de différences dans l'écologie, mais également dans la génétique des écotypes de mêmes populations. Ces constats supportent donc l'idée que le polymorphisme associé aux ressources engendrerait, dans certains cas, un isolement reproducteur (voir Hendry 2009, Pfennig et al. 2010 pour des exemples). Bien que le polymorphisme associé aux ressources ne soit pas toujours associé à un isolement reproducteur ou à des différences génétiques entre les écotypes, ces phénomènes pourraient permettre à la sélection naturelle (dans ce cas-ci souvent divergente) d'amplifier les différences entre les écotypes.

Des exemples de polymorphisme associé aux ressources ont été répertoriés chez de nombreuses espèces, et ce, au sein d'ordres variés (Smith et Skúlason 1996). Par exemple, le pyréneste ponceau (Pyrenestes ostrinus), un oiseau de la famille des estrildidés, présente un dimorphisme au niveau de la grosseur du bec (Smith 1990) où un écotype possédant

un bec de grande taille semble mieux adapté à se nourrir des graines de carex (dures) alors qu'un écotype doté d'un petit bec se nourrirait plus efficacement de graines molles. Chez les amphibiens, les têtards du crapaud à couteaux du Nouveau-Mexique (*Spea multiplicata*) présentent un dimorphisme associé aux ressources alimentaires (formes carnivore et omnivore) se différenciant par la longueur de l'intestin et la grosseur des muscles hyoïdiens orbitaux (Pfennig 1992).

Chez les poissons, le polymorphisme associé aux ressources se traduit très souvent par l'existence de formes littorale et pélagique (Robinson et Wilson 1994). Cette dichotomie est explicable par la présence de deux habitats distincts, les zones littorales et pélagiques, qui diffèrent par leur structure physique et leurs ressources alimentaires dans les lacs de l'hémisphère nord (Skúlason et Smith 1995). De plus, il est possible de placer les différentes populations ou espèces qui expriment un polymorphisme associé aux ressources le long d'un continuum de spéciation sympatrique (Hendry 2009). En effet, à l'une des extrémités de ce gradient, nous retrouvons des exemples de polymorphisme très contrasté, où les différences morphologiques et écologiques entre les écotypes sont très importantes. Ces exemples de polymorphisme peuvent également être marqués par un isolement reproducteur important entre les écotypes, phénomène primordial pour qu'une population polymorphique puisse accomplir une spéciation complète. L'omble chevalier (*Salvelinus alpinus*) est un exemple typique de polymorphisme (contrasté) pouvant présenter de deux (Hindar et Jonsson 1982, Knudsen et al. 2010) à quatre écotypes différents à l'intérieur d'un même lac (Malmquist et al. 1992, Snorrason et al. 1994) et présenter un isolement reproducteur contraignant le flux génétique entre les écotypes (Wilson et al. 2004). À l'autre extrême de ce continuum, nous retrouvons des populations ou espèces présentant une diversification phénotypique beaucoup plus subtile entre les écotypes (Hendry 2009). Le crapet-soleil (*Lepomis gibbosus*) et l'omble de fontaine (*Salvelinus fontinalis*) en sont de bons exemples, car les différences morphologiques ne peuvent être détectées que par des analyses statistiques, n'étant pas apparentes à l'examen visuel des individus (Robinson et Wilson 1994, Bourke et al. 1997, Dynes et al. 1999, Proulx et Magnan 2002, Bertrand et al. 2008). Chez ces espèces, il a été démontré que le polymorphisme associé aux ressources possède des composantes génétique et phénotypique (Robinson et Wilson

1994, Proulx et Magnan 2004, Wilson et McLaughlin 2007). Cependant, leur importance respective semble varier non seulement entre les espèces, mais également entre les populations (Smith et Skúlason 1996). Ainsi, pour mieux comprendre les mécanismes menant à la spéciation sympatrique, l'étude des facteurs influençant l'expression du polymorphisme associé aux ressources semble un incontournable, et ce, pour différentes espèces se situant le long du continuum de spéciation.

1.3 Processus évolutifs menant au polymorphisme

L'identification des mécanismes sous-jacents aux processus de diversification n'est pas un objectif nouveau en écologie et a même été étudiée dans « On the Origin of Species » (Darwin 1859). Pourtant, de par leur nature et l'échelle à laquelle ils opèrent, l'identification des facteurs favorisant la diversification est encore à ce jour un défi de taille en écologie évolutive (Pfennig et al. 2010; Wellborn et Langerhans 2015; Skúlason et al. 2019). La plupart des théories supposent que le point de départ des processus de diversification menant au polymorphisme associé aux ressources serait l'apparition d'opportunités écologiques (Yoder et al. 2010; Skúlason et al. 2019). Les opportunités écologiques se présentent lorsque de nouvelles ressources deviennent disponibles suite à la colonisation d'un nouvel habitat, à l'extinction de compétiteurs/prédateurs qui accaparaient ou limitaient, auparavant, une partie des ressources disponibles ou encore à la suite de l'apparition d'innovations clés (c.-à-d., nouvelle adaptation) permettant l'exploitation de nouvelles ressources (Yoder et al. 2010). Lorsqu'une opportunité écologique se présente, il est généralement reconnu que c'est la pression de compétition intraspécifique qui pousse certains individus à se tourner vers ces nouvelles ressources (peu utilisées) pour échapper aux fortes pressions de cette compétition (Bolnick 2001). Cette augmentation de l'étendue totale de la niche de la population engendrerait également une augmentation des variations phénotypiques (morphologiques et comportementales) en association à l'exploitation de nouvelles ressources, un phénomène appelé relâchement écologique (english : « *ecological release* ») (Schluter 2000; Yoder et al. 2010). Ainsi, le terme relâchement écologique correspond aux réponses d'une population en lien à l'exploitation d'une opportunité écologique. Ce terme regroupe donc plusieurs

phénomènes souvent interreliés englobant notamment la relaxation de la sélection naturelle (Roughgarden, 1972), l'élargissement de l'étendue totale des ressources et des habitats utilisés (Bolnick et al. 2010), une augmentation de la densité des individus de ladite population (MacArthur et al., 1972) ainsi que l'augmentation de la variation phénotypique (Nosil et Reimchen, 2005).

1.4 Contexte et objectifs de la thèse

Considérant que les lacs postglaciaires (âgés de 10 000 – 15 000 ans) dimictiques de l'hémisphère nord affichent deux habitats bien distincts (zones littorale et pélagique), ces derniers semblent propices au polymorphisme associé aux ressources (Schluter et McPhail 1993, Robinson et Wilson 1994, Skúlason et al. 2019). C'est notamment le cas de l'omble de fontaine, qui présente un polymorphisme subtil associé aux ressources dans certains lacs du Bouclier laurentien : un écotype littoral présente des nageoires pectorales plus longues, une forme plus trapue et s'alimente de proies benthiques dans la zone littorale alors qu'un autre écotype pélagique présente des nageoires pectorales plus courtes, un aspect plus fusiforme et s'alimente de zooplancton dans la colonne d'eau (Bourke et al. 1997, 1999; Dynes et al. 1999, Marchand et al. 2002, Proulx et Magnan 2002). Des nageoires longues sont associées à une meilleure manœuvrabilité, favorisant une alimentation d'organismes benthiques retrouvés sur le substrat de la zone littorale, alors que des nageoires pectorales plus courtes et une forme du corps plus fusiforme sont associées à une nage de type « croisière », requise dans la recherche de proies comme le zooplancton en eau libre (zone pélagique) (Webb 1984, Ehlinger 1990). Une autre étude a montré que la faune parasitaire des individus littoraux et pélagiques était caractéristique des hôtes intermédiaires associés aux zones littorale et pélagique respectivement, suggérant que ce polymorphisme est stable dans le temps (Bertrand et al. 2008). De plus, la morphologie des poissons capturés dans les deux zones était fortement corrélée à la faune parasitaire, suggérant une diversification fonctionnelle des deux groupes. Rouleau et al. (2010) ont quant à eux observé que les individus pélagiques issus de parents provenant du milieu naturel ont affiché de meilleures performances de nage que les individus littoraux, suggérant que les différences morphologiques ont une valeur sélective

et fonctionnelle. Ces auteurs ont également observé que les individus provenant de lignées hybrides avaient des performances de nage inférieures aux lignées littorale et pélagique, ce qui supporte l'hypothèse de la valeur sélective associée aux écotypes spécialisés (Schluter 1995). Il a également été montré que certains comportements associés à l'alimentation (ex. : taux de captures et nombre de proies rejetées) étaient transmissibles des parents indigènes, provenant d'un lac, à leur progéniture, élevée en laboratoire (Sacotte et Magnan 2006). De plus, certains traits morphologiques étaient sous influence environnementale (ex. : longueur de la mandibule inférieure), et même réversible, alors que d'autres se sont avérés être sous une influence génétique (ex. : longueur des nageoires pectorales et dorsales) (Proulx et Magnan 2004, Sacotte et Magnan 2006). Des travaux échelonnés de 2000 à 2007 sur la frayère du lac Ledoux (Réserve Mastigouche, Québec) ont permis de découvrir que les individus de la forme littorale (basés sur la morphologie) arrivent majoritairement sur la frayère dans les deux premières semaines de la saison de reproduction et les individus pélagiques, dans les deux dernières semaines, suggérant un début d'isolement reproducteur entre les deux formes (Magnan et al. en prép.). Finalement, il a été montré que la population d'omble de fontaine du lac Ledoux (Réserve Mastigouche, Québec) possédait deux grandes tactiques thermiques, dont une « tempérée » (eaux peu profondes) et une « froide » (eaux profondes) (Bertolo et al. 2011, Goyer et al. 2014). Même si ces tactiques n'ont pu être associées formellement aux formes littorale et pélagique, les auteurs ont suggéré qu'il s'agissait de l'explication la plus parcimonieuse. Dans ce contexte, les individus littoraux seraient exposés à un bilan thermique estival plus élevé, qui pourrait entraîner une maturation des gonades et des caractères sexuels secondaires plus hâtive, les poussant à se rendre plus tôt sur la frayère, ce qui pourrait entraîner un certain isolement reproducteur (Magnan et al., en prép.). Or, le polymorphisme associé aux ressources exprimé par l'omble de fontaine a été étudié sur un nombre très restreint de lacs et s'est avéré variable d'une population à l'autre. De plus, plusieurs espèces de poisson ont été introduites par l'homme au cours du dernier siècle dans ce système et leur effet, en tant que force de compétition interspécifique, sur l'expression du polymorphisme associé aux ressources chez l'omble de fontaine n'a pas été étudiés à ce jour. On retrouve trois niveaux de compétition interspécifique associés aux espèces introduites dans ce système : les lacs où l'omble de fontaine est la seule espèce

présente (c.-à-d., sans compétition interspécifique), des lacs où il vit en sympatrie avec le mulet à cornes, *Semotilus atromaculatus* (c.-à-d., compétition interspécifique intermédiaire), et des lacs où il vit également le meunier noir, *Catostomus commersonii* (c.-à-d., compétition interspécifique forte) (Bourke et al. 1999).

Le premier objectif de cette thèse (Chapitre II) a donc été de déterminer l'influence de la compétition intra- et interspécifique ainsi que de plusieurs facteurs environnementaux (c.-à-d., la température de l'épilimnion, la proportion de la zone littorale et la saisonnalité) sur l'utilisation des habitats littoral et pélagique par l'omble de fontaine. Pour ce faire, nous avons d'abord évalué l'effet de ces différents facteurs sur l'abondance des ombles de fontaine dans les habitats littoral et pélagique indépendamment. Puis, nous y développons un cadre théorique basé sur les isodars (Morris 1988). Les isodars permettent entre autres de déterminer le ou les habitats préférés par l'espèce cible, si cette préférence est dépendante de la densité et comment la compétition interspécifique et les facteurs environnementaux affectent les patrons spatiaux d'abondance de l'espèce cible dans chacun des habitats. Comme les espèces présentes dans ce système (omble de fontaine, mulet à cornes et meunier noir) semblent partager le même habitat préférentiel (zone littorale) et que le meunier noir est un compétiteur supérieur dans cet habitat (Magnan 1988, Bourke et al. 1999), nous prédisons que plus l'abondance relative des compétiteurs sera élevée dans un lac, plus l'omble de fontaine sélectionnera l'habitat pélagique. Aussi, suivant la même logique, plus la densité en omble de fontaine sera élevée dans les lacs allopatriques, plus la proportion d'individus sélectionnant la zone pélagique sera élevée.

L'objectif du chapitre III a été de tester l'hypothèse de l'opportunité écologique dans ce système. Une opportunité écologique se présente lorsqu'une ressource devient disponible à la suite du relâchement de la compétition interspécifique ou de la prédation, menant à la colonisation de cette niche vacante par une autre espèce (Schluter 2000, Yoder et al. 2010). Il est généralement suggéré que la plasticité phénotypique et la compétition intraspécifique sont les principaux facteurs induisant cette diversification de l'utilisation des ressources et que ces phénomènes seraient les principaux responsables de l'expression du polymorphisme associé aux ressources (Martin et Pfennig 2010, Nosil et Reimchen

2005). Pour ce faire, nous avons évalué le degré de spécialisation alimentaire entre les écotypes pélagique et littoral au sein des différents niveaux de compétition interspécifique précédemment décrits, à l'aide de cinq indicateurs de l'utilisation des ressources (contenus stomacaux, $\delta^{13}C$ du foie, concentration en caroténoïdes, longueur des ceaca pyloriques et des branchicténies). Selon l'hypothèse de l'opportunité écologique, nous devrions observer une plus grande spécialisation entre des écotypes dans les lacs où la compétition interspécifique est faible. De plus, le régime alimentaire de l'écotype littoral devrait davantage s'apparenter à celui de l'écotype pélagique dans les lacs ou la compétition interspécifique est forte en zone littorale.

L'objectif du chapitre IV a été de vérifier, à une grande échelle (18 populations), quels sont les traits morphologiques impliqués dans la spécialisation aux habitats littoral et pélagique des deux écotypes et de vérifier si différentes populations présentaient une évolution parallèle des deux écotypes. L'évolution parallèle d'écotypes prend place lorsque les écotypes de populations distinctes présentent des adaptations similaires en réponse à des environnements équivalents (Schluter 2000). Dans ce chapitre, nous avons également évalué l'impact de la compétition interspécifique en zone littorale sur la morphologie des ombles de fontaine littoraux et pélagiques. Pour ce faire, nous avons comparé la forme du corps ainsi que 17 traits morphologiques entre les écotypes littoral et pélagique de 18 populations d'omble de fontaine. Cette approche permet notamment de déterminer la présence d'évolution parallèle le long d'un gradient de compétition interspécifique et de déterminer si ces populations subissent des forces de sélection divergentes similaires.

En plus d'acquérir des connaissances fondamentales sur la dynamique des populations de l'omble de fontaine en milieu lacustre, les travaux menés dans le cadre de cette thèse auront certainement une incidence importante en écologie fonctionnelle et évolutive. En effet, l'approche à une grande échelle (c.-à-d., l'étude de près d'une trentaine de populations) que nous proposons, a permis de généraliser et d'améliorer la compréhension théorique des mécanismes responsables de l'expression du polymorphisme associé aux ressources chez l'omble de fontaine lacustre du Bouclier laurentien.

CHAPITRE II

INTRASPECIFIC COMPETITION AND TEMPERATURE DRIVE HABITAT-BASED RESOURCE POLYMORPHISM IN BROOK CHARR, *SALVELINUS FONTINALIS*, WITH SPECIAL REFERENCE TO CLIMATE CHANGES

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Author contributions

PM and AD originally formulated the idea. VR and MP conducted fieldwork. VR wrote the first draft of the manuscript. All authors were involved in data analyses, contributed critically to the drafts, and gave final approval for publication.

Abstract

Intra- and interspecific competition are the two main driving forces suggested to promote habitat-based resource polymorphism, but very few studies have provided empirical support for this hypothesis. Furthermore, although competition has been extensively studied in density-dependent habitat selection, the constraints of external drivers are often omitted when studying animal distribution among habitats. Specifically, ambient temperature is known to affect the fitness of ectotherms and may have important consequences on habitat-based resource polymorphism. Using mixed-effects modelling and isodar analyses, we quantified the effects of water temperature and habitat availability as well as intra- and interspecific competition on the abundance and density-dependent habitat selection of two brook charr (*Salvelinus fontinalis*) between littoral and pelagic habitats, in 27 Canadian Shield lakes. We found that high interspecific competition by white sucker (*Catostomus commersonii*) drastically reduced brook charr abundances in both the littoral and pelagic habitats but did not affect their density-dependent habitat selection. In contrast, the isodar analysis showed density-dependent habitat selection, confirming that intraspecific competition is a driver of their habitat use. Furthermore, brook charr preferred the littoral habitat at water temperatures below 20.8°C, but they preferred the pelagic habitat above this threshold and no longer used the littoral habitat when its temperature was above ~22.2°C. By incorporating an external driver in the density-dependent habitat selection model, we show how the littoral temperature can shape distribution patterns between the littoral and pelagic habitats, i.e., by acting as a thermal barrier and thus limiting the willingness of fish to use resources in the littoral habitat. This result suggests that global warming could restrain the diversifying effect of intraspecific competition and prevent resource polymorphism, a phenomenon promoting adaptive radiation and speciation.

Keywords: density dependence, habitat selection, isodar, salmonids, thermal preference, behavioural thermoregulation.

Introduction

Resource polymorphism, i.e., when discrete phenotypes of the same population show differential niche use, may represent an important intermediate stage in sympatric speciation (Smith and Skúlason 1996; Schluter 2000; Skúlason et al. 2019). Common examples of this phenomenon are fish species found in depauperate post-glacial lakes of the northern hemisphere (Smith and Skúlason 1996). These lakes almost always include coexisting benthic and pelagic ecotypes that are better adapted to feeding on bottom organisms or zooplankton, respectively (Robinson and Wilson 1994; Smith and Skúlason 1996). A first factor promoting resource polymorphism would be the exploitation of a new or unexploited resource to reduce intraspecific competition through phenotypic plasticity and divergent selection (selection against intermediates) (Smith and Skúlason 1996; Svanbäck and Bolnick 2007; Martin and Pfennig 2010; Araújo et al. 2011; Skúlason et al. 2019). A second factor would be release from interspecific competition (Smith and Skúlason 1996; Svanbäck and Bolnick 2007; Martin and Pfennig 2010), referred to as ecological opportunity. Yet very few studies have provided evidence that intra- and interspecific competition are driving forces of resource polymorphism.

When considering the effect of intraspecific competition (i.e., density dependence) in habitat selection, the ideal free distribution (IFD) theory proposes that individuals maximize their fitness by selecting the most suitable habitats (Fretwell and Lucas 1969). However, as the density of organisms competing for the same resources increases in a habitat, its resource availability and thus its suitability often decrease relative to alternative habitats (which had initially been less suitable). In these conditions, IFD theory states that organisms will distribute among habitats in a way that the fitness of all individuals will be equal (Fretwell and Lucas 1969). This basic principle has been used to developed isodar theory (Morris 1988), which allows the inference of drivers of habitat selection based only on abundance patterns. The habitat isodar is the regression line between the density of individuals in one habitat and their density in an adjacent habitat, for which the fitness of individuals is equalized (Morris 1988). When different from zero, the isodar intercept indicates the difference in the maximum potential fitness that can be attained in each habitat at low density due to differences in resource quantity (Morris 1988). The slope of

the isodar provides information on the relative rate at which fitness decreases with increasing density in each habitat; more directly, it reveals the influence of any density-dependent process (intraspecific competition or density-dependent predation) on habitat selection (Morris 1988). A slope that is not significantly different from zero indicates that habitat selection is not affected by intraspecific competition. A slope of one indicates that density-dependent processes have the same influence on fitness in the two habitats while a slope significantly different from one indicates differences between habitats that would affect resource quality (such as its acquisition) differently in the two habitats at high and low population densities (Morris 1988). Thus, an isodar with an intercept of 0 and a slope of 1 indicates that the two habitats are similar and have the same influence on fitness. This approach can also be used to assess the influence of interspecific competition (e.g., Rodríguez 1995), predation (e.g., Dupuch et al. 2014), and environmental factors like temperature (e.g., Halliday and Blouin-Demers 2017) on density-dependent habitat selection. In this context, the isodar theory could be a strong conceptual framework to address the driving forces of habitat-based resource polymorphism.

Brook charr, *Salvelinus fontinalis,* exhibit a subtle resource polymorphism in some Canadian Shield lakes (Bourke et al. 1997; Dynes et al. 1999; Proulx and Magnan 2002; Proulx and Magnan 2004; Bertrand et al. 2008). The littoral ecotype has a deeper body and longer pectoral fins, is found in shallow water (0-2 m), and feeds mainly on zoobenthos, while the pelagic ecotype has a more streamlined body shape with shorter pectoral fins, is found in deeper waters (3-6 m), and feeds mostly on zooplankton. Morphological differences between ecotypes seem to be determined by both genetic and environmental factors (Dynes et al. 1999; Proulx and Magnan 2004; Sacotte and Magnan 2006) and are functionally related to the swimming performance and energetics of the two forms (Peres-Neto and Magnan 2004; Rouleau et al. 2010).

During the last century, bait fishers introduced creek chub (*Semotilus atromaculatus*) and white sucker (*Catostomus commersonii*) to many Canadian Shields lakes. Creek chub and white sucker compete for food with brook charr in the littoral zone (Magnan 1988; Lachance and Magnan 1990a; Lachance and Magnan 1990b; Tremblay and Magnan 1991;

Lacasse and Magnan 1992) and decrease its relative abundance and biomass by 30% to 70% (Magnan 1988; Lachance and Magnan 1990a; Magnan et al 2005), suggesting a strong impact of interspecific competition of introduced species in the littoral zone. In these lakes, the level of interspecific competition varies from none (allopatric brook charr lakes), to intermediate (brook charr with creek chub lakes), and to high (brook charr with creek chub and white sucker lakes) (Magnan 1988; Bourke et al. 1999).

Finally, elevated summer temperatures could prevent brook charr from foraging in the littoral zone while refuging in the cool water layer of the metalimnion in the pelagic zone (Goyer et al. 2014). Many cold-water fish such as brook charr use behavioural thermoregulatory tactics to access certain resources while minimizing the negative impacts of suboptimal temperature (Sims et al. 2006; Bertolo et al. 2011; Goyer et al. 2014). Thus, water temperature in the littoral zone may also be an important driver of brook charr habitat selection.

We used the isodar theory and abundance data of brook charr in the littoral and pelagic habitats of 27 Canadian Shield lakes (along the gradient of interspecific competition described above) to determine the influence of intra- and interspecific competition as well as higher water temperature in the littoral zone on brook charr habitat selection. Isodar analyses are mainly based on the density of individuals in contrasting habitats (Morris 1988), but terms that have an influence on habitat quality can be added into models (Hodson et al. 2010). We developed a framework based on the isodar theory that illustrates how higher water temperature in the littoral zone can influence density-dependent habitat selection of an ectotherm species using behavioural thermoregulation.

Theoretical framework

If intraspecific competition influences habitat selection of brook charr, the fitness of individuals will decrease with an increasing abundance of fish in the littoral and pelagic habitats (Fig. 1a and 1c), and their abundance in the preferred habitat (the littoral zone, N_L) will be influenced by the abundance of fish in the pelagic habitat (N_P; Fig. 1b and 1d),

i.e., $N_L = \beta_0 + \beta_1 N_P$. Brook charr prefer to feed in the littoral habitat, based on its feeding habits in allopatry (Lacasse and Magnan 1992; Bourke et al. 1999); this may be due to higher quantities of food resources and thus to a higher maximum fitness that can be attained at low brook charr density in this habitat relative to the pelagic one (Fig. 1a). If so, this will produce a significant isodar intercept ($\beta_0 > 0$; Fig. 1b). This preference could also result from a lower influence of brook charr density on individual fitness in the littoral compared to the pelagic habitat (Fig. 1c), in which case it would be associated with an isodar slope greater than one ($\beta_1 > 1$; Fig. 1d). It is known that in ectotherms, such as cold-water fish, higher water temperature in the littoral zone (T_L) can reduce this habitat's thermal quality through a negative impact on individual fitness. As shown in Halliday and Blouin-Demers (2018), such a negative effect on fitness occurs when temperature deviates from the optimal temperature, either because the thermal reaction norm drives the maximum fitness that can be achieved by an individual in the littoral zone (i.e., the isodar intercept is reduced as temperature deviates from the optimal temperature) or because the energetic demand (i.e., demand for food resources) or food acquisition (i.e., the isodar slope) decrease through a reduced metabolic rate as temperature deviates from the optimum. If so, the negative density dependence of individual fitness will be reduced in the littoral habitat and the isodar slope will increase (figure not provided; e.g., Halliday and Blouin-Demers 2017). However, brook charr use thermoregulatory behaviour to maintain body temperature near the thermal optimum (Bertolo et al. 2011; Goyer et al. 2014). This species has been observed to significantly decrease their selected temperatures and daily movements in the littoral habitat when the mean daily epilimnion temperature is above 22.4°C (Goyer et al. 2014). Consequently, higher water temperature in the littoral zone is unlikely to influence brook charr fitness through an alteration in body temperature (i.e., through an alteration of the isodar intercept; $N_L = \beta_0 + \beta_1 N_P + \beta_2 T_L$, with $\beta_2 < 0$; Fig. 1[e-f]). However, higher water temperature can reduce brook charr food acquisition in the littoral zone by reducing the time they can spend in this habitat (Pépino et al. 2018). Furthermore, short excursions likely increase inter-habitat movements and so the per capita demand on food resources (due to increased energetic demand). These two potential effects ultimately increase the negative density dependence of fitness, especially in the preferred littoral habitat (Fig. 1g), and the isodar slope will decrease as water temperature

increases in this habitat ($N_L = \beta_0 + \beta_1 N_P + \beta_3 T_L \times N_P$, with $\beta_3 < 0$; Fig. 1h). We used this conceptual framework to evaluate the response of brook charr to the increased water temperature occurring in the littoral zone during summer.

Materials and Methods

Study area and fish sampling

The study was conducted in Mastigouche (46° 40′ N, 73° 30′ W) and Saint-Maurice (47° 05′ N, 73° 15′ W) wildlife reserves and in La Mauricie National Park (46°45′ N, 73°08′ W), Québec, Canada, from June to August in 2012, 2013, and 2014. We sampled 10 lakes containing only brook charr, eight with brook charr and creek chub, and nine with brook charr, creek chub, and white sucker, hereafter referred to as brook charr (BC), brook charr and creek chub (BC+CC), and brook charr, creek chub, and white sucker (BC+CC+WS) lakes (Table 1). Other fish species were found in BC+CC and BC+CC+WS lakes (Table 1), but the main competitors of brook charr in this system are creek chub and white sucker (Magnan 1988; Magnan et al. 2005). Fish were caught in both the littoral (< 3 m) and pelagic (> 4 m) zones of the lakes using four trap nets (Alaska type; opening 1.0 m × 1.8 m, equipped with two 1 m × 15 m wings; Fipec Industries, Gaspé, Québec, Canada). The traps were randomly located in each zone and fished for four (2013-2014) or five (2012) days from 18:00 to 6:00 to capture brook charr at the time that they are most active in their feeding habitat (based on previous studies in this system; Bourke et al. 1996; Bertolo et al. 2011; Goyer et al. 2014). Each brook charr was identified by clipping a small part of the left pelvic fin and released. Recaptured individuals (< 0.6% of the total brook charr captured) were not considered in the CPUEs. We estimated the water temperature for each trap net using thermograph lines installed at the deepest location of each lake.

Statistical analyses

Mixed effects modelling of brook charr abundance

We first analyzed brook charr abundance in littoral and pelagic habitats independently to determine the effect of interspecific competition and water temperature on brook charr abundance in each zone. In addition, we examined three other factors that may influence habitat and food availability for brook charr: the relative proportions of the littoral and pelagic habitats as well as the day and year of sampling. The relative proportions of the littoral and pelagic habitats influence the relative carrying capacity of each habitat in each lake and thus their availability for brook charr (see Bertrand et al. 2008). The day of sampling is associated with the abundance of zoobenthic communities, which decreases throughout the summer in comparable temperate or boreal lakes (D; Mittelbach 1981; Persson 1987). Food resources are also expected to fluctuate among years; day and year of sampling are therefore two variables that can affect food availability and potentially habitat use by brook charr. The decrease of benthic organisms throughout the summer season is compatible with seasonal variations in brook charr feeding habits in our system (Lacasse and Magnan 1992; Chapter III). To this end, we used two generalized linear mixed models with negative binomial distributions (function glmer.nb; lme4, v1.1-21; Bates et al. 2015) to fit our dependent variables, i.e., number of brook charr catch per trap (catch per unit of effort; CPUE) in littoral and pelagic habitats. The mixed effects modelling approach allowed us to account for the hierarchical structure of the data, with brook charr CPUE nested within lake. These models (Table 2) included different combinations of the following predictive variables of interest: 1) Water temperature at trap depth; 2) Intensity of interspecific competition (categorical variable: none [brook charr only], intermediate [brook charr and creek chub], and high [brook charr, creek chub, and white sucker]); 3) the proportion of littoral habitat area relative to lake area; 4) day of year; and 5) sampling year ($Year_1$ and $Year_2$ variables were included as categorical covariates, coded as (-1, -1), (1, 0), and (0, 1) for years 2012, 2013, and 2014, respectively).

We used Akaike Information Criterion adjusted for small sample size, AICc (Burnham and Anderson 2002), to select the best model. Models were ranked using ΔAICc, i.e., the difference in AICc between a candidate model and the model with the lowest (best) AICc. We selected the model with the lowest AICc score, except when several models were within two units of the lowest scoring model, in which case we selected the most parsimonious model (i.e., with the smallest number of parameters).

Density-dependent habitat selection using isodar analysis

This first set of analyses allowed us to identify three key variables that influence brook charr abundance in littoral and pelagic habitats, i.e., interspecific competition, water temperature in the littoral zone, and day of year (see results). However, the structure of the above models did not take intraspecific competition into account. We tested the effect of intraspecific competition in combination with water temperature in the littoral zone (Fig. 1) and the two other key variables using the isodar theory. Linear regression (function lm; stats, v3.5.2; R Development Core Team 2015) was used to assess the different isodars included in the conceptual framework (Fig. 1; models 0, 4, and 7 in Table 3). The intensity of interspecific competition © was entered as an additional predictive variable into the isodar through an effect on the isodar intercept ($N_L = \beta_0 + \beta_1 N_P + \beta_4 C$; model 2 in Table 3) or slope ($N_L = \beta_0 + \beta_1 N_P + \beta_5 C \times N_P$; model 8 in Table 3) to represent exploitation competition (i.e., a reduction in available food quantity due to the presence of interspecific competitors, which decreases the isodar intercept, $\beta_4 < 0$) or interference competition (i.e., a reduction in food acquisition due to the presence of interspecific competitors, which decreases the isodar slope, $\beta_5 < 0$), respectively. Because brook charr abundance modelled for each habitat independently revealed no significant difference between no interspecific competition and intermediate levels of interspecific competition, we reduced interspecific competition to two levels – with and without white sucker – for the isodar models. We also included a day of year effect (D) in the isodar intercept ($N_L = \beta_0 + \beta_1 N_P + \beta_6 D$; model 1 in Table 3) to quantify the effect of the progressive seasonal decline in zoobenthic community abundance (i.e., food availability) on brook charr density-dependent habitat selection.

We built 16 isodars predicting the abundance of brook charr in the littoral habitat (N_L) with different combinations of the explanatory variables (Table 3). These candidate isodars were limited to a maximum of three explanatory terms because of the small dataset (N = 27). The best isodar was selected based on AICc following the procedure described above. We inspected residuals from the isodar models for non-linearity (Knight and Morris 1996), but none was apparent within the range of observed brook charr abundances. We then estimated equation coefficients for the best isodar model using geometric mean regression based on the standard minor axis method (McArdle 1988; Richter and Stavn 2014). We calculated the "bias-corrected and accelerated" (Bca) 95% confidence intervals for each estimate included in the selected model using bootstrapping techniques (boot package: n = 1000; Efron and Tibshirani 1994). This method is more appropriate to fit a line to a bivariate dataset where errors are present in both variables and when we want to make inferences on the intercept and the slope of the relationship (McArdle 1988).

Results

Brook charr abundance in littoral and pelagic habitats

Two models predicting brook charr abundances in both the littoral and pelagic habitats fall under the threshold of two ΔAICc units (models 5 and 7 in each habitat; Table 2). The first and most parsimonious model includes the terms for water temperature at the trap depth (T) and intensity of interspecific competition © in both habitat models. These models predict comparable abundances of brook charr with no interspecific competition and with intermediate levels of interspecific competition, but much less in lakes with high competition (Fig. 2a and 3a, respectively). These models also predict that brook charr abundance will decrease in the littoral habitat and increase in the pelagic habitat as littoral water temperature increases (Fig. 2b and 3b, respectively) and as summer progresses, independently of water temperature (Tables 4 and 5, respectively). The second set of models also includes the proportion of littoral habitat (P_L) and predicts that brook charr abundance will be higher in both the littoral and pelagic habitats in lakes where the littoral habitat is relatively more important (Tables 6 and 7, respectively).

Density-dependent habitat selection using isodar analysis

The most parsimonious isodar model (model 7; Table 3) includes the relative abundance of brook charr in the pelagic habitat (N_P) and its interaction with the mean littoral temperature ($T \times N_P$). The relative abundance of brook charr in the littoral habitat was positively related to its abundance in the pelagic habitat (N_P estimate; $\beta_1 = 15.75$; 95% CI = 12.77, 22.59), and this relationship decreased as water temperature in the littoral zone increased ($T \times N_P$ estimate; $\beta_3 = -0.71$; 95% CI = -1.09, -0.58). The isodar intercept was not significantly different from zero (intercept; $\beta_0 = 0.84$; 95% CI = -5.22, 7.23). When the littoral water temperature was under 20.8°C, the isodar revealed a clear density-dependent preference of brook charr for the littoral habitat. However, this preference started to shift to the pelagic habitat as the littoral water temperature increased above 20.8°C, i.e., the temperature at which the littoral and pelagic habitats are similar (slope 1:1) (Fig. 4). This model thus reveals a strong effect of the littoral water temperature on brook charr density-dependent habitat selection.

Discussion

Our study addressed the impacts of intra- and interspecific competition and littoral water temperature on lacustrine brook charr habitat selection between the littoral and pelagic zones. In agreement with our predictions, both high water temperature and interspecific competition (i.e., lakes with white sucker) reduced brook charr abundance in the littoral habitat. Furthermore, the overall increase of brook charr abundance with an increased proportion of littoral habitat highlights the importance of the littoral habitat in the overall fish carrying capacity of these lakes (e.g., Biró and Vörös 1990; Taylor et al. 2020). Our analysis also revealed density-dependent habitat selection by brook charr between littoral and pelagic habitats, with individuals preferring the littoral habitat when water temperature was below 20.8°C (slope > 1). However, the preference started to shift to the pelagic zone (slope < 1:1) when temperatures rose to between 20.8°C and 22.2°C, with 22.2°C being the temperature at which brook charr habitat selection is no longer density dependent (isodar slope = 0). The increase in water temperature may thus have

important ecological and evolutionary impacts on resource polymorphism in cold-water species.

Intra- and interspecific competition

This study showed density-dependent habitat selection between littoral and pelagic habitats. This finding confirms that intraspecific competition is involved in brook charr habitat use, which is one the main factors hypothesized to promote resource polymorphism through phenotypic plasticity and divergent selection (selection against intermediates) (Smith and Skúlason 1996; Svanbäck and Bolnick 2007; Martin and Pfennig 2010; Araújo et al. 2011; Skúlason et al. 2019). The possible mechanism behind this implies that as intraspecific pressure increases (via increased population density or resource depletion), the initially suboptimal habitat becomes more profitable (Bolnick 2001). Therefore, individuals using suboptimal resources will experience decreased intraspecific competition and so their fitness will be equal to those individuals using preferred resources (Roughgarden 1972). For instance, using lake enclosures, Svanbäck and Bolnick (2007) showed that increasing the density of three-spine stickleback populations caused individuals of different phenotypes to add different alternative prey types to their diet, suggesting that distinct ecotypes will shift to distinct habitats with increasing density in natural systems. Relative to intraspecific competition, interspecific competition did not have a significant effect on habitat selection for brook charr in our system. Brook charr populations subjected to creek chub and white sucker competition incorporate more pelagic resources into their diet (Bourke et al. 1999). In this study, we did not find the expected negative impacts of competition by creek chub on brook charr abundance or habitat selection. However, competition by white sucker drastically reduced brook charr abundances by 71% and 65% in littoral and pelagic, respectively, but did not influence their density-dependent habitat selection. These results suggest that, in our system, intraspecific rather than interspecific competition is the main driver of lacustrine brook charr resource polymorphism.

Seasonality and temperature

Our results also showed that, independently of littoral water temperature, brook charr abundance in the littoral habitat decreases as the summer progresses, suggesting a habitat shift to the pelagic zone as the abundance of zoobenthic communities declines through the summer (Mittelbach 1981; Persson 1983, 1986, 1987; Tremblay and Magnan 1991). This interpretation is further supported by our observation that the opposite phenomenon takes place in the pelagic habitat: brook charr abundances tended to increase as the summer progressed. The effect of seasonality is also well captured by the isodar approach, where the day of the year explained 52% of density-dependent habitat selection.

Beyond seasonality, higher water temperature in the littoral zone reduces the isodar slope (interaction term between brook charr abundance in the pelagic habitat and water temperature in the selected model), suggesting that higher water temperature does not influence the maximum fitness that an individual can achieve in the littoral habitat (through the thermal reaction norm, i.e., the isodar intercept), but increases the negative density-dependence of fitness (through higher energetic demand and/or reduced food acquisition) in this habitat. Halliday and Bouin-Demers (2014) suggested that when thermal temperature deviates from the optimal temperature, the ability to process resources (not to acquire resources) should be the rate-limiting factor. In contrast, our results suggest that for species using thermoregulatory behaviour to maintain optimal body temperature in a heterogeneous thermal environment, warming temperatures induce fitness costs mainly through energetic demand and food acquisition. The isodar analysis also suggested that when the littoral water temperature exceeds 22.2°C, brook charr habitat selection is no longer density dependent and individuals completely cease to venture into the littoral habitat. Using radio telemetry, Goyer et al. (2014) observed that brook trout significantly decreased their daily movements in the epilimnion when its mean daily temperature was above 22.4°C; this value closely matches the upper temperature threshold published for this species (Wismer and Christie 1987; Smith and Ridgway 2019). In a study simulating body temperature in a heterogeneous thermal environment, Pépino et al. (2015) suggested that excursions between warm and cold temperatures, defined as behavioural thermoregulation (Bertolo et al. 2011), could be adopted by brook

trout to keep their body temperature below the critical temperature threshold, enabling them to exploit habitats with temperatures higher than this threshold for short periods of time. In this context, the 22.2°C threshold observed in the present study, as well as the value of 22.4°C observed by Goyer et al. (2014), probably corresponds to the temperature where it is no longer possible for brook charr to use littoral resources without suffering the negative consequences of harmful body temperatures. Therefore, the increase in brook charr abundances in the pelagic habitat as the littoral temperature warms is probably a consequence of the individual's decision to no longer use the littoral habitat and to seek thermal refuge in the pelagic habitat. In this context, the phenological diversity of food resources (Armstrong et al. 2016) as well as the spatial arrangement of thermal refuges through time (Sears et al. 2016) will be of primary importance in predicting potential behavioural adaptations of cold-stenothermic fish species to warming environments (White et al. 2019).

Lake warming

Finally, recent projections have estimated that lake surface-water temperatures will rise by several degrees during the current century in response to climate change (O'Reilly et al. 2015; Woolway and Merchant 2019). Loss of transparency is also expected to accentuate the effects of global warming by reducing epilimnion thickness, increasing the thermal gradient between surface and bottom waters, and prolonging the stratification period, which could lead to deoxygenation of the cool, deep waters (Bartosiewicz et al. 2019). It is possible that ongoing modifications of lake thermal regimes could occur too rapidly, and fish species could be prevented specializing on specific environments if evolutionary processes are slower than environmental change. In this context, we suggest that the behavioural plasticity of habitat use – and especially behavioural thermoregulation – could buffer cold-water fish species against climate warming as proposed for terrestrial ectotherms (Kearney et al. 2009; Sunday et al. 2014). Our study showed that high littoral water temperatures affect brook charr habitat selection to the point that the littoral habitat will not be used when temperatures are above a critical threshold, with potential consequences on fish growth and condition (e.g., Guzzo et al. 2017). Thus, behavioural

thermoregulation may not always have the expected buffering effect against climate warming. Understanding thermal tolerance and its effect on behavioural thermoregulation, as well as the acclimation capacity of distinct populations and how these characteristics will change according to ontogeny and in varying environments (in space and time), will improve our ability to quantify the real impacts of climate change on the fitness of fish populations (Stitt et al. 2014; Sinclair et al. 2016). Overall, our results provide insight into the potential impact of global warming on cold-water species because it reduces their willingness to use littoral food resources during the summer period and consequently, their ability to use behavioural thermoregulation against global warming. Ultimately, this reduces the role of intraspecific competition (i.e., density-dependence) in habitat selection of the two brook charr ecotypes between the littoral and pelagic habitats, i.e., the main driving force behind habitat-based resource polymorphism in this species. In an evolutionary perspective, global warming could restrain the diversifying effect of intraspecific competition and prevent resource polymorphism, a phenomenon promoting adaptive radiation and speciation.

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References

Araújo MS, Bolnick DI, Layman CA (2011) The ecological causes of individual specialisation. Ecology Letters 14: 948-958. Doi: 10.1111/j.1461-0248.2011.01662.x

Armstrong JB, Takimoto G, Schindler DE, Hayes MM, Kauffman MJ (2016) Resource waves: phenological diversity enhances foraging opportunities for mobile consumers. Ecology 97: 1099-1112. Doi: 10.1890/15-0554.1

Bartosiewicz M, Przytulska A, Lapierre J-F, Laurion I, Lehmann MF, Maranger R (2019) Hot tops, cold bottoms: synergistic climate warming and shielding effect increase carbon burial in lakes. Limnology and Oceanography Letters 4: 132-144. Doi: 10.1002/lol2.10117

Bates D, Maechler M, Bolker B, Walker S (2015) Fitting linear mixed-effects models using lme4. Journal of Statistical Software, 67: 1-48. Doi: 10.18637/jss.v067.i01

Bertolo A, Pépino M, Adams J, Magnan P (2011) Behavioural thermoregulatory tactics in lacustrine brook charr, *Salvelinus fontinalis*. PloS ONE. 6: e18603. Doi: 10.1371/journal.pone.0018603

Bertrand M, Marcogliese DJ, Magnan P (2008) Trophic polymorphism in brook charr revealed by diet, parasites and morphometrics. Journal of Fish Biology 72: 555-572. Doi:10.1111/j.1095-8649.2007.01720.x

Biró P, Vörös L (1990) Trophic relationships between primary producers and fish yields in Lake Balaton. Hydrobiologia 191: 213-221.

Bolnick DI (2001) Intraspecific competition favours niche width expansion in *Drosophila melanogaster*. Nature 410: 463-466.

Bourke P, Magnan P, Rodriguez MA (1996) Diel locomotor activity of brook charr, as determined by radiotelemetry. Journal of Fish Biology 49: 1174-1185.

Bourke P, Magnan P, Rodriguez MA (1997) Individual variations in habitat use and morphology in brook charr. Journal of Fish Biology 51: 783-794.

Bourke P, Magnan P, Rodriguez MA (1999) Phenotypic responses of lacustrine brook charr in relation to the intensity of interspecific competition. Evolutionary Ecology 13: 19-31.

Burnham KP, Anderson DR (2002) Model selection and multimodel inference: A practical information-theoretic approach. Springer-Verlag, New York.

Dupuch A, Morris DW, Ale SB, Wilson DJ, Moore DE (2014) Landscapes of fear or competition? Predation did not alter habitat choice by Arctic rodents. Oecologia 174: 403-412.

Dynes J, Magnan P, Bernatchez L, Rodriguez MA (1999) Genetic and morphological variation between two forms of lacustrine brook charr. Journal of Fish Biology 54: 955-972.

Efron B, Tibshirani RJ (1994) An introduction to the bootstrap (monographs on statistics and applied probability). Chapman and Hall, London.

Fretwell SD, Lucas HL (1969) On territorial behavior and other factors influencing habitat distributions of birds: I. Theoretical development. Acta Biotheoretica 19: 16-36.

Goyer K, Bertolo A, Pépino M, Magnan P (2014) Effects of lake warming on behavioural thermoregulatory tactics in a cold-water stenothermic fish. PloS ONE 9: e92514. Doi: 10.1371/journal.pone.0092514

Guzzo MM, Blanchfield PJ, Rennie MD (2017) Behavioral responses to annual temperature variation alter the dominant energy pathway, growth, and condition of a cold-water predator. Proceedings of the National Academy of Sciences 114: 9912-9917. Doi: 0.1073/pnas.1702584114

Halliday WD, Blouin-Demers G (2014) Red flour beetles balance thermoregulation and food acquisition via density-dependent habitat selection. Journal of Zoology 294: 198–205. Doi: 10.1111/jzo.12168

Halliday WD, Blouin-Demers G (2017) Can temperature modify the strength of density-dependent habitat selection in ectotherms? A test with red flour beetles. Journal of Zoology 304: 159-168. Doi: 10.1111/jzo.12510

Halliday WD, Blouin-Demers G (2018) Body temperature influences growth rates of Common Gartersnakes (*Thamnophis sirtalis*). Canadian Field-Naturalist 132: 25-29. Doi: 10.22621/cfn.v132i1.2018

Hodson J, Fortin D, LeBlanc ML, Belanger L (2010) An appraisal of the fitness consequences of forest disturbance for wildlife using habitat selection theory. Oecologia 164: 73-86. Doi: 10.1007/s00442-010-1691-4

Kearney M, Shine R, Porter WP (2009) The potential for behavioral thermoregulation to buffer "cold-blooded" animals against climate warming. Proceedings of the National Academy of Sciences of the United States of America 106: 3835-3840. Doi: 10.1073/pnas.0808913106

Knight TW, Morris DW (1996) How many habitats do landscapes contain? Ecology 77: 1756-1764.

Lachance S, Magnan P (1990a) Performance of domestic, hybrid, and wild strains of brook trout, *Salvelinus fontinalis*, after stocking: the impact of intra- and interspecific competition. Canadian Journal of Fisheries and Aquatic Sciences 47: 2278-2284.

Lachance S, Magnan P (1990b) Comparative ecology and behavior of domestic, hybrid, and wild strains of brook trout, *Salvelinus fontinalis*, after stocking. Canadian Journal of Fisheries and Aquatic Sciences 47: 2285-2292.

Lacasse S, Magnan P (1992) Biotic and abiotic determinants of the diet of brook trout, *Salvelinus fontinalis*, in lakes of the Laurentian Shield. Canadian Journal of Fisheries and Aquatic Sciences 49: 1001-1009.

Magnan P (1988) Interaction between brook charr, *Salvelinus fontinalis*, and nonsalmonid species: Ecological shift, morphological shift, and their impact on zooplankton communities. Canadian Journal of Fisheries and Aquatic Sciences 45: 999-1009.

Magnan P, Proulx R, Plante M (2005) Integrating the effects of fish exploitation and interspecific competition into current life history theories: an example with lacustrine brook trout (*Salvelinus fontinalis*) populations. Canadian Journal of Fisheries and Aquatic Sciences 62: 747-757. Doi: 10.1139/F05-041

Martin RA, Pfennig DW (2010) Field and experimental evidence that competition and ecological opportunity promote resource polymorphism. Biological Journal of the Linnean Society 100: 73-88. Doi: 10.1111/j.1095-8312.2010.01380.x

McArdle BH (1988) The structural relationship: regression in biology. Canadian Journal of Zoology 66: 2329-2339.

Mittelbach GG (1981) Patterns of invertebrate size and abundance in aquatic habitats. Canadian Journal of Fisheries and Aquatic Sciences 38: 896-904.

Morris DW (1988) Habitat-dependent population regulation and community structure. Evolutionary Ecology 2: 253-269.

O'Reilly CM, Sharma S, Gray DK, Hampton SE, Read JS, Rowley RJ et al. (2015) Rapid and highly variable warming of lake surface waters around the globe. Geophysical Research Letters 42: 10773-10781. Doi: 10.1002/2015GL066235

Pépino M, Goyer K, Magnan P (2015) Heat transfer in fish: are short excursions between habitats a thermoregulatory behaviour to exploit resources in an unfavourable thermal environment? Journal of Experimental Biology 218: 3461-3467. Doi:10.1242/jeb.126466

Pépino M, Magnan P, Proulx R (2018) Field evidence for a rapid adaptive plastic response in morphology and growth of littoral and pelagic brook charr: A reciprocal transplant experiment. Functional Ecology 32: 161-170. Doi: 10.1111/1365-2435.12929

Peres-Neto PR, Magnan P (2004) The influence of swimming demand on phenotypic plasticity and morphological integration: a comparison of two polymorphic charr species. Oecologia 140: 36-45. Doi: 10.1007/s00442-004-1562-y

Persson L (1983) Food consumption and competition between age classes in a perch, *Perca fluviatilis*, population in a shallow eutrophic lake. Oikos 40: 197-207.

Persson L (1986) Effects of reduced interspecific competition on resource utilization in perch (*Perca fluviatilis*). Ecology 67: 355-364.

Persson L (1987) The effects of resource availability and distribution on size class interactions in perch, *Perca fluviatilis*. Oikos 48: 148-160.

Proulx R, Magnan P (2002) Physiological performance of two forms of lacustrine brook charr, *Salvelinus fontinalis*, in the open-water habitat. Environmental Biology of Fishes 64: 127-136.

Proulx R, Magnan P (2004) Contribution of phenotypic plasticity and heredity to the trophic polymorphism of lacustrine brook charr (*Salvelinus fontinalis* M.). Evolutionary Ecology Research 6: 503-522.

R Development Core Team (2015) R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. (n.d.). http://www.r-project.org.

Richter SJ, Stavn RH (2014) Determining functional relations in multivariate oceanographic systems: Model II multiple linear regression. Journal of Atmospheric and Oceanic Technology 31: 1663-1672. Doi: 10.1175/JTECH-D-13-00210.1

Robinson BW, Wilson DS (1994) Character release and displacement in fishes: a neglected literature. The American Naturalist 144: 596-627.

Rodríguez MA (1995) Habitat-specific estimates of competition in stream salmonids: A field test of the isodar model of habitat selection. Evolutionary Ecology 9: 169-184.

Roughgarden J (1972) Evolution of niche width. The American Naturalist 106: 683-718.

Rouleau S, Glémet H, Magnan P (2010) Effects of morphology on swimming performance in wild and laboratory crosses of brook trout ecotypes. Functional Ecology 24: 310-321. Doi: 10.1111/j.1365-2435.2009.01636.x

Sacotte S, Magnan P (2006) Inherited differences in foraging behaviour in the offspring of two forms of lacustrine brook charr. Evolutionary Ecology Research 8: 843-857.

Sears MW, Angilletta MJ, Schuler MS, Borchert J, Dilliplane KF, Stegman M, Rusch TW, Mitchell WA (2016) Configuration of the thermal landscape determines thermoregulatory performance of ectotherms. Proceedings of the National Academy of Sciences 113: 10595-10600. Doi: 10.1073/pnas.1604824113

Schluter D (2000) The ecology of adaptive radiation. Oxford Univ. Press, Oxford.

Sims DW, Wearmouth VJ, Southall EJ, Hill JM, Moore P, Rawlinson K, Hutchinson N, Budd GC, Righton D, Metcalfe JD, Nash JP, Morritt D (2006) Hunt warm, rest cool: bioenergetic strategy underlying diel vertical migration of a benthic shark. Journal of Animal Ecology 75: 176-190. Doi: 10.1111/j.1365-2656.2005.01033.x

Sinclair BJ, Marshall KE, Sewell MA, Levesque DL, Willett CS, Slotsbo S, Dong Y, Harley CDG, Marshall DJ, Helmuth BS, Huey RB (2016) Can we predict ectotherm responses to climate change using thermal performance curves and body temperatures? Ecology Letters 19: 1372-1385. Doi: 10.1111/ele.12686

Skúlason S, Parsons KJ, Svanbäck R, Räsänen K, Ferguson MM, Adams CE, Amundsen PA, Bartels P, Bean CW, Boughman JW, Englund G, Guðbrandsson J, Hooker OE, Hudson AG, Kahilainen KK, Knudsen R, Kristjánsson BK, Leblanc CAL, Jónsson Z, Öhlund G, Smith C, Snorrason SS (2019) A way forward with eco evo devo: an extended theory of resource polymorphism with postglacial fishes as model systems. Biological Reviews 94: 1786-1808. Doi: 10.1111/brv.12534

Smith DA, Ridgway MS (2019) Temperature selection in Brook Charr: lab experiments, field studies, and matching the Fry curve. Hydrobiologia, 840: 143-156. Doi: 10.1007/s10750-018-3869-4

Smith TB, Skúlason S (1996) Evolutionary significance of resource polymorphisms in fishes, amphibians, and birds. Annual Review of Ecology and Systematics 27: 111-133.

Stitt BC, Burness G, Burgomaster KA, Currie S, McDermid JL, Wilson CC (2014) Intraspecific variation in thermal tolerance and acclimation capacity in Brook Trout (*Salvelinus fontinalis*): physiological implications for climate change. Physiological and Biochemical Zoology 87: 15-29. Doi: 10.1086/675259

Sunday JM, Bates AE, Kearney MR, Colwell RK, Dulvy NK, Longino JT, Huey RB (2014) Thermal-safety margins and the necessity of thermoregulatory behavior across latitude and elevation. Proceedings of the National Academy of Sciences 111: 5610-5615. Doi: 10.1073/pnas.1316145111

Svanbäck R, Bolnick DI (2007) Intraspecific competition drives increased resource use diversity within a natural population. Proceedings of the Royal Society B 274: 839-844. Doi: 10.1098/rspb.2006.0198

Taylor TN, Cross BK, Moore BC (2020) Modeling brook trout carrying capacity in Owhi Lake, Washington, using bioenergetics. North American Journal of Fisheries Management 40: 84-104. Doi: 10.1002/nafm.10378

Tremblay S, Magnan P (1991) Interactions between two distantly related species, brook trout (*Salvelinus fontinalis*) and white sucker (*Catostomus commersoni*). Canadian Journal of Fisheries and Aquatic Sciences. 48: 857-867.

White SL, Kline BC, Hitt NP, Wagner T (2019) Individual behaviour and resource use of thermally stressed brook trout *Salvelinus fontinalis* portend the conservation potential of thermal refugia. Journal of Fish Biology 95: 1061-1071. Doi: 10.1111/jfb.14099

Wismer DA, Christie AE (1987) Temperature relationships of Great Lakes fishes: A data compilation. Great Lakes Fishery Commission Special Publication No. 87-3. Ann Arbor, Michigan.

Woolway RL, Merchant CJ (2019) Worldwide alteration of lake mixing regimes in response to climate change. Nature Geoscience 12: 271-276. Doi: 10.1038/s41561-019-0322-x

Tables

Table 1. Characteristics of the 27 studied brook charr lakes. Only creek chub and white sucker were included when determining the mean competitor biomass.

Lake	Sampling week date	Fish community*	Area (ha)	Mean depth (m)	Littoral zone area proportion (%)	Littoral zone volume proportion (%)	Mean competitor biomass in littoral (g/trap)
Baie des onze îles (A)	19-Jun + 26-Jul-2012		135	7.20	18	7	0
Baie Verte (B)	26-Jun + 1-Aug-2012		11	5.04	51	38	0
Baie Cobb (C)	03 + 20-Jul-2012		62	5.88	34	8	0
Maréchal (D)	10 + 17-Jul-2012		103	5.18	42	10	0
Vertnez (E)	20-May-2013	SEAT	-	-	-	-	16
Bucheron (G)	03-Jun-2013		10	3.90	41	18	0
Cutaway (H)	10-Jun-2013	CHEO, SEAT, CACO	40	3.38	52	15	312
Camp (I)	17-Jun-2013	CHEO, SEAT, CACO	11	5.82	44	13	505
Visons (J)	24-Jun-2013	CHEO, SEAT	74	7.54	31	6	12
Jimmy (K)	02-Jul-2013	CHEO, SEAT, CACO, LUCO	-	-	-	-	169
Brise (L)	08-Jul-2013	CHEO, SEAT, CACO	39	3.59	63	23	304
Lézard (M)	15-Jul-2013	CHEO, SEAT, CACO	110	7.83	24	5	469
Chute noire (N)	22-Jul-2013	CHEO	17	3.80	45	18	0
Corneille (O)	29-Jul-2013	CHEO, SEAT, CACO	23	4.78	48	17	347
Bourassa (P)	05-Aug-2013	CHEO	56	7.26	28	8	0
Coteau (Q)	20-May-2014	CHEO	31	4.72	43	16	0
Oudiette (R)	27-May-2014	CHEO, SEAT, CACO	44	8.59	27	7	211

Table 1 (Continued)

Lake	Sampling week date	Fish community*	Area (ha)	Mean depth (m)	Littoral zone area proportion (%)	Littoral zone volume proportion (%)	Mean competitor biomass in littoral (g/trap)
Gauthier (S)	03-Jun-2014	CHEO, SEAT, MAMA	37	8.62	35	9	22
Shawinigan (T)	10-Jun-2014	CHEO, SEAT, LUCO, MAMA	60	11.70	18	4	26
Adam (U)	17-Jun-2014		14	5.24	42	13	0
Dorval (V)	24-Jun-2014	CHEO, CACO, COPL, CUIN, RHCA	-	-	-	-	285
Marshall (W)	01-Jul-2014	SEAT	41	6.39	33	10	48
Marmotte (X)	08-Jul-2014	SEAT	14	2.97	50	27	38
Longue vue (Y)	15-Jul-2014	SEAT	28	5.44	45	16	18
Plouf (Z)	22-Jul-2014	CHEO, SEAT, CACO, PIPR	60	8.82	29	5	378
Thibert (AA)	29-Jul-2014	SEAT	43	5.79	36	8	28
Simpson (BB)	05-Aug-2014		30	4.24	45	26	0

* Fish species codes are SEAT, creek chub (*Semotilus atromaculatus*); CHEO, northern redbelly dace (*Chrosomus eos*); CACO, white sucker (*Catostomus commersonii*); LUCO, common shiner (*Luxilus cornutus*); MAMA, Allegheny pearl dace (*Margariscus margarita*); COPL, lake chub (*Couesius plumbeus*); CUIN, brook stickleback (*Culaea inconstans*); RHCA, longnose dace (*Rhinichthys cataractae*); and PIPR, fathead minnow (*Pimephales promelas*). Brook charr (*Salvelinus fontinalis*) was present in all lakes.

Table 2. Comparison of all candidate models predicting the relative abundance of brook charr (CPUE) in littoral and pelagic habitats. Number of model parameters (K) and difference in AICc relative to the best-fitting model (ΔAICc) are presented. Model terms are T, water temperature at the trap depth (°C); C, interspecific competition intensity (none, low, high); and P_L, proportion of littoral habitat (area). Each model also includes the standardized (i.e., $\mu = 0$, $\sigma = 1$) day of year as a fixed effect. The sampling year ($Year_1$ and $Year_2$) variables were included as categorical covariates, coded as (-1, -1), (1, 0), and (0, 1) for 2012, 2013, and 2014, respectively. Lake was included as a random effect in all models.

Model	Terms in the model	K	ΔAICc Littoral Habitat	ΔAICc Pelagic Habitat
0	D	6	10.5	16.5
1	T + D	**7**	4.9	8.3
2	P_L + D	7	9.9	18.2
3	C + D	8	5.7	5.2
4	T + P_L + D	8	5.3	8.5
5	**T + C + D**	**9**	**0.5**	**0.7**
6	P_L + C + D	9	4.1	6.4
7	T + C + P_L + D	10	0.0	0.0

Table 3. Comparison of all isodar candidate models predicting the relative abundance of brook charr (CPUE) in the littoral habitat (N_L). Number of model parameters (K), difference in AICc relative to the best-fitting model (ΔAICc), and R-squared adjusted for the number of predictors are presented. Model terms are N_P, brook charr relative abundance in the pelagic habitat (CPUE); T_L, mean littoral water temperature (°C); C, interspecific competition (without or with white sucker); and D, day of year. These terms are included to model their effect on resource quantity (i.e., effects on isodar intercept; models 1 to 6), on resource acquisition (i.e., effects on isodar slope; models 7 to 9), or both (models 10 to 15).

Model		K	ΔAICc	R^2_{adj}
Intraspecific competition only				
0	$N_L = N_P$	3	26.9	0.00
Resource quantity				
Effect of seasonality				
1	$N_L = N_P + D$	4	8.4	0.52
Effect of interspecific competition (exploitation)				
2	$N_L = N_P + C$	4	19.2	0.28
Effect of interspecific competition and seasonality				
3	$N_L = N_P + C + D$	5	3.1	0.63
Effect of littoral water temperature				
4	$N_L = N_P + T_L$	4	5.5	0.57
5	$N_L = N_P + T_L + D$	5	6.2	0.58
6	$N_L = N_P + T_L + C$	5	2.0	0.64
Resource acquisition				
Effect of littoral water temperature (harmful water temperature)				
7	$\mathbf{N_L = N_P + T_L \times N_P}$	**4**	**1.9**	**0.62**
Effect of interspecific competition (interference)				
8	$N_L = N_P + C \times N_P$	4	26.6	0.05
Effect of littoral water temperature and interspecific competition				
9	$N_L = N_P + C \times N_P + T_L \times N_P$	5	3.7	0.63
Resource acquisition and quantity				
10	$N_L = N_P + C \times N_P + D$	5	10.8	0.51
11	$N_L = N_P + C \times N_P + C$	5	21.5	0.27
12	$N_L = N_P + C \times N_P + T_L$	5	7.3	0.57
13	$N_L = N_P + T_L \times N_P + D$	5	3.5	0.62
14	$N_L = N_P + T_L \times N_P + C$	5	0.0	0.67
15	$N_L = N_P + T_L \times N_P + T_L$	5	2.8	0.64

Table 4. Estimates of the selected competing model (model #5; Table 2) predicting relative abundance in the littoral habitat based on 251 traps in 27 lakes. This model is based on a negative binomial distribution with a shape parameter (θ) of 1.33. Initial day-of-the-year (D) distribution was 188.7 ± 21.9 days. Initial water temperature (T) distribution was 20.4 ± 2.8°C. In this model, D and T were standardized (i.e., $\mu = 0$, $\sigma = 1$). C: Interspecific competition intensity (three-level categorical variable: none [brook charr only], intermediate [brook charr and creek chub], and high [brook charr, creek chub, and white sucker]); BC is the reference category. The sampling year ($Year_1$ and $Year_2$) variables were included as categorical covariates, coded as (-1, -1), (1, 0), and (0, 1) for 2012, 2013, and 2014, respectively.

Random effects	**Variance**	**Std. Dev.**
Lake (Intercept)	0.60	0.78

Fixed effects	**Estimate**	**Std. Error**	**z value**	**Pr (>|z|)**
Intercept	2.61	0.27	9.82	< 0.001
T	-0.44	0.16	-2.69	0.037
C: BC+CC	-0.09	0.50	-0.18	0.794
C: BC+CC+WS	-1.47	0.48	-3.05	0.003
D	-0.61	0.19	-3.20	0.002
$Year_1$	-0.12	0.28	-0.43	0.948
$Year_2$	0.41	0.28	1.44	0.112

Table 5. Estimates of the selected competing model (model #5; Table 2) predicting relative abundance in the pelagic habitat based on 217 traps from 27 lakes. This model is based on a negative binomial distribution with a shape parameter (θ) of 1.94. Initial day-of-the-year (D) distribution was 183.7 ± 22.0 days. Initial water temperature at trap depth (T) distribution was 9.8 ± 3.7°C. In this model, D and T were standardized (i.e., $\mu = 0$, $\sigma = 1$). C: Interspecific competition intensity (three-level categorical variable: none [brook charr only], intermediate [brook charr and creek chub], and high [brook charr, creek chub, and white sucker]); BC is the reference category. The sampling year ($Year_1$ and $Year_2$) variables were included as categorical covariates, coded as (-1, -1), (1, 0), and (0, 1) for 2012, 2013, and 2014, respectively.

Random effects	**Variance**		**Std. Dev.**	
Lake (Intercept)	0.15		0.39	
Fixed effects	**Estimate**	**Std. Error**	**z value**	**Pr (>|z|)**
Intercept	2.60	0.15	17.50	< 0.001
T	0.24	0.09	2.53	0.011
C: BC+CC	0.09	0.27	0.32	0.747
C : BC+CC+WS	-0.92	0.27	-3.45	< 0.001
D	0.15	0.10	1.46	0.144
$Year_1$	-0.43	0.16	-2.77	0.006
$Year_2$	0.23	0.16	1.48	0.140

Table 6. Estimates of the best competing model (model #7; Table 2) predicting relative abundance in the littoral habitat based on 251 traps in 27 lakes. This model is based on a negative binomial distribution with a shape parameter (θ) of 1.33. Initial day-of-the-year (D) distribution was 188.7 ± 21.9 days. Initial water temperature (T) distribution was 20.4 ± 2.8°C. Initial distribution of the proportion of littoral habitat (P_{Litt}) was 0.40 ± 0.10. In this model, P_{Litt} values were log transformed ($\log_e(x)$). D, T, and P_{Litt} were standardized (i.e., $\mu = 0$, $\sigma = 1$). C: Interspecific competition intensity (three-level categorical variable: none [brook charr only], intermediate [brook charr and creek chub], and high [brook charr, creek chub, and white sucker]); BC is the reference category. The sampling year ($Year_1$ and $Year_2$) variables were included as categorical covariates, coded as (-1, -1), (1, 0), and (0, 1) for 2012, 2013, and 2014, respectively.

Random effects	**Variance**	**Std. Dev.**
Lake (Intercept)	0.54	0.74

Fixed effects	**Estimate**	**Std. Error**	**z value**	**Pr (>|z|)**
Intercept	2.57	0.26	10.02	< 0.001
T	-0.40	0.16	-2.46	0.014
C: BC+CC	-0.03	0.48	-0.07	0.948
C: BC+CC+WS	-1.44	0.46	-3.11	0.002
P_{Litt}	0.28	0.17	1.61	0.108
D	-0.63	0.19	-3.40	< 0.001
$Year_1$	-0.17	0.27	-0.60	0.547
$Year_2$	0.47	0.27	1.72	0.086

Table 7. Estimates of the best competing model (model #7; Table 2) predicting relative abundance in the pelagic habitat based on 217 traps from 27 lakes. This model is based on a negative binomial distribution with a shape parameter (θ) of 1.95. Initial day-of-the-year (D) distribution was 183.7 ± 22.0 days. Initial water temperature at trap depth (T) distribution was 9.8 ± 3.7°C. Initial distribution of the proportion of littoral habitat (P_{Litt}) was 0.40 ± 0.10. In this model, P_{Litt} values were log transformed ($\log_e(x)$) and D, T, and P_{Litt} were standardized (i.e., $\mu = 0$, $\sigma = 1$). C: Interspecific competition intensity (three-level categorical variable: none [brook charr only], intermediate [brook charr and creek chub], and high [brook charr, creek chub, and white sucker]); BC is the reference category. The sampling year ($Year_1$ and $Year_2$) variables were included as categorical covariates, coded as (-1, -1), (1, 0), and (0, 1) for 2012, 2013, and 2014, respectively.

Random effects	**Variance**		**Std. Dev.**	
Lake (Intercept)	0.13		0.37	
Fixed effects	**Estimate**	**Std. Error**	**z value**	**Pr (>|z|)**
Intercept	2.57	0.15	17.71	< 0.001
T	0.27	0.09	2.89	0.004
C: BC+CC	0.17	0.27	0.62	0.537
C : BC+CC+WS	-0.89	0.26	-3.46	< 0.001
P_{Litt}	0.17	0.10	1.71	0.087
D	0.13	0.10	1.30	0.194
$Year_1$	-0.45	0.15	-3.01	0.003
$Year_2$	0.28	0.16	1.76	0.079

Figures captions

Figure 1. Conceptual framework based on isodar theory. Four scenarios of expected fitness (W)-density (N) functions (left panels) and corresponding isodars (right panels) for littoral (red) and pelagic (blue) habitats when water temperature in the littoral zone is optimal (solid black and red lines; from a to d) and when it becomes harmful (dashed black and red lines; from e to h). The grey dashed lines represent the 1:1 slope (see text for the different assumptions). Scenario 1: at optimal temperature, brook charr preference for the littoral habitat is due to a higher quantity of food resources in this habitat relative to the pelagic habitat (a); this condition is associated with a significant isodar intercept (b) ($\beta_0 > 0$). Scenario 2: a lower influence of brook charr density on individual fitness in the littoral compared to the pelagic habitat (c), which is associated with an isodar slope higher than one (d) ($\beta_1 > 1$). Assuming that preference for the littoral habitat is associated with scenario 2, when temperature deviates from the optimal value in the littoral habitat, we expect scenario 3: a reduction of the maximum fitness achieved by an individual in the littoral zone © and the associated reduction of isodar intercept (f), or scenario 4: the combined effect of behavioural thermoregulatory costs (increased energetic demand) and shorter excursions into the littoral habitat that may intensify the negative effect of density on fitness in this habitat (g) and in turn decrease isodar slope as littoral temperature increases (h).

Figure 2. Partial residual plot (estimate ± 95% CI) for a) fish community © at 20.35°C (mean temperature) and b) water temperature at the trap depth (T). Fish community: BC: brook charr lakes (blue); BC+CC: brook charr and creek chub lakes (orange); and BC+CC+WS: brook charr, creek chub, and white sucker lakes (red). The dashed grey line represents the mean temperature (20.22°C).

Figure 3. Partial residual plot (estimate ± 95% CI) for a) fish community © at 9.81°C (mean temperature) and b) water temperature at the trap depth (T). Fish community: BC: brook charr lakes (blue); BC+CC: brook charr and creek chub lakes (orange); and

BC+CC+WS: brook charr, creek chub, and white sucker lakes (red). The dashed grey line represents the mean temperature (9.78°C).

Figure 4. Habitat isodars for brook charr in littoral (Y axis) and pelagic (X axis) habitats in relation to littoral water temperature. Fitted lines from the best model II regression are shown from 11°C (dark blue) to 22°C (red) with 1°C interval between each line. The mean relative abundance in littoral and pelagic habitats are presented for BC and BC+CC lakes (filled circles) and BC+CC+WS lakes (filled triangles). The grey dashed line represents the 1:1 slope.

Figures

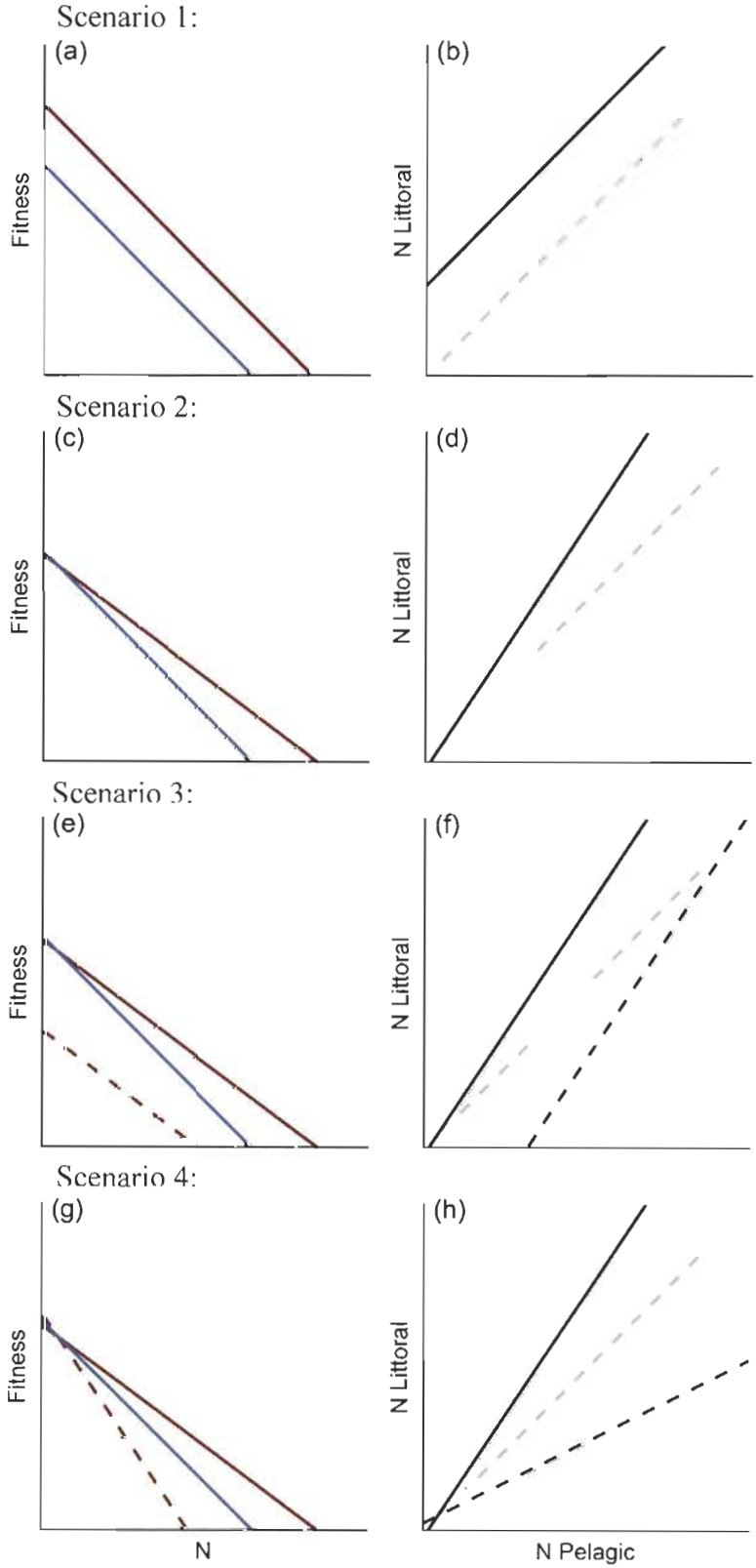

Figure 1.

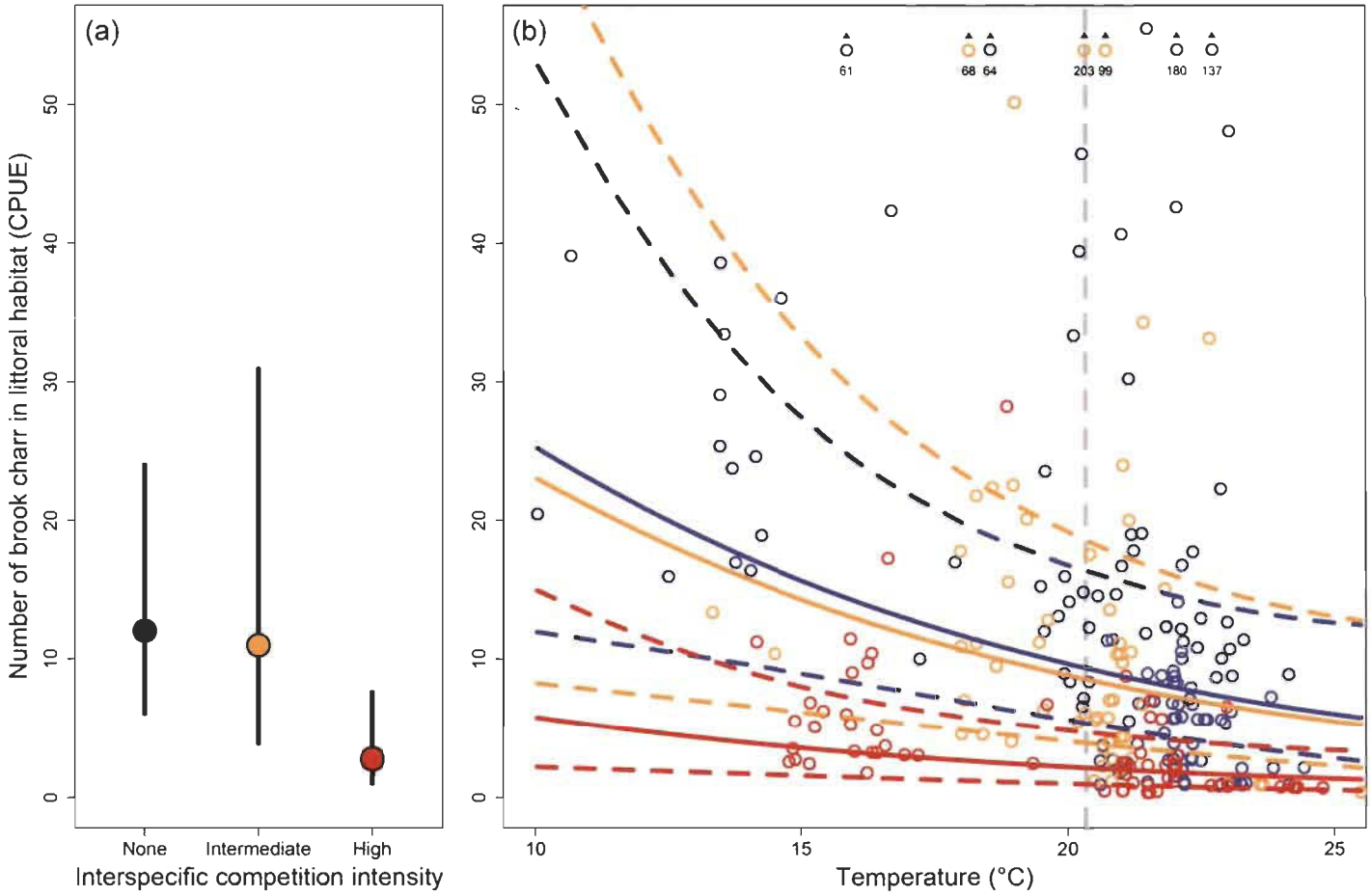
(a)
Number of brook charr in littoral habitat (CPUE)
0
10
20
30
40
50
None
Intermediate
High
Interspecific competition intensity
(b)
61
68 64
203 99
180 137
0
10
20
30
40
50
10
15
20
25
Temperature (°C)

Figure 2.

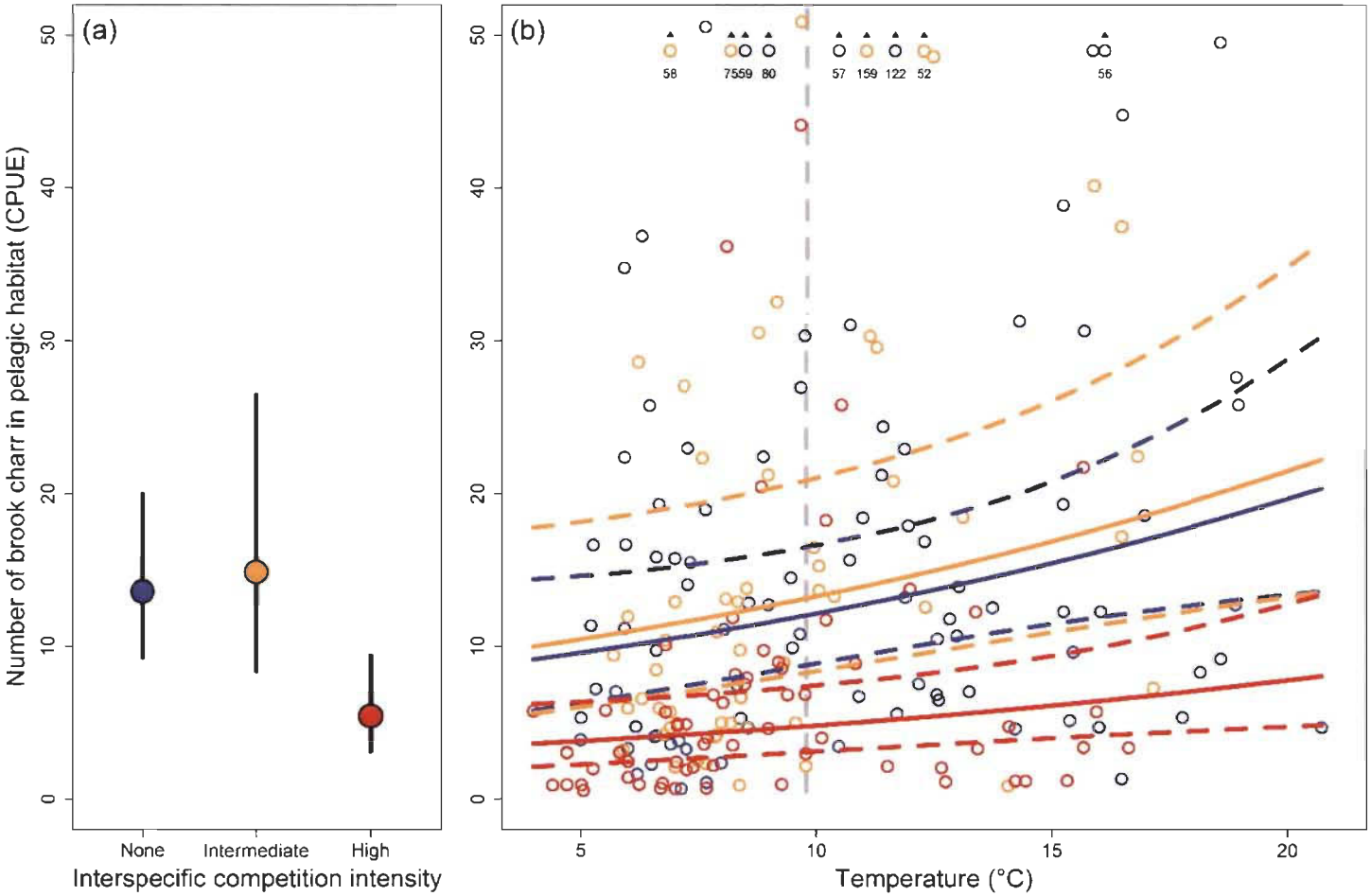
(a)
Number of brook charr in pelagic habitat (CPUE)
0
10
20
30
40
50
None
Intermediate
High
Interspecific competition intensity
(b)
58
75
59
80
57
159
122
52
56
0
10
20
30
40
50
5
10
15
20
Temperature (°C)

Figure 3.

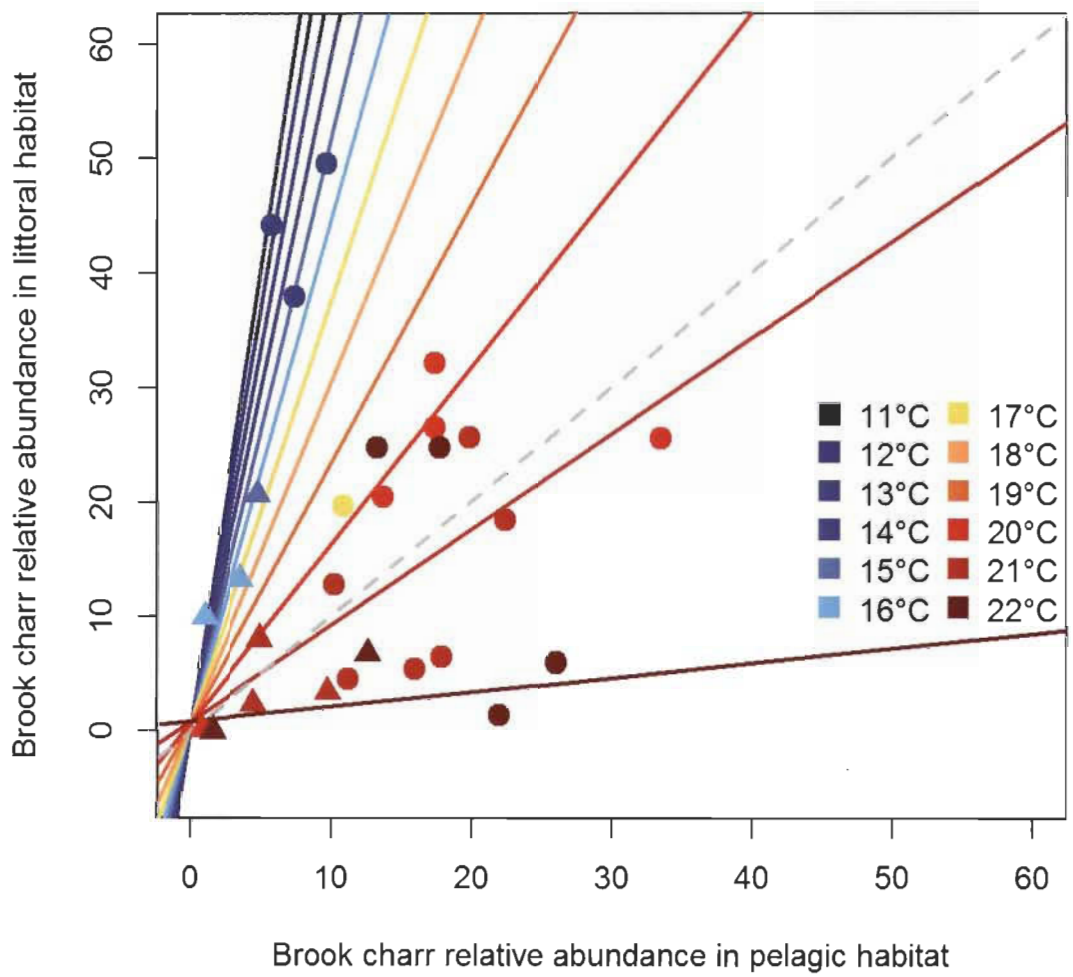
Brook charr relative abundance in littoral habitat
0
10
20
30
40
50
60
Brook charr relative abundance in pelagic habitat
0
10
20
30
40
50
60
11°C
12°C
13°C
14°C
15°C
16°C
17°C
18°C
19°C
20°C
21°C
22°C

Figure 4.

CHAPITRE III

DOES ECOLOGICAL RELEASE FROM DISTANTLY RELATED SPECIES AFFECT PHENOTYPIC DIVERGENCE IN BROOK CHARR?

Cet article est sous presse dans le périodique *Oecologia*.

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Author contributions

PM originally formulated the idea. VR, MP, IL, and AF conducted fieldwork. VR, IL, and AF conducted laboratory analyses. VR wrote the first draft of the manuscript. All authors were involved in data analyses, contributed critically to the drafts, and gave final approval for publication.

Abstract

Ecological opportunity occurs when a resource becomes available through a decrease of interspecific competition and another species colonizes the vacant niche through phenotypic plasticity and intraspecific competition. This hypothesis has been supported in many studies involving closely related species, but very few studies have examined distantly related lineages. Brook charr (Salvelinus fontinalis) exhibit a resource polymorphism in some Canadian Shield lakes, where a littoral ecotype feeds mainly on zoobenthos and a pelagic ecotype feeds mostly on zooplankton. Creek chub (*Semotilus atromaculatus*) and white sucker (*Catostomus commersonii*), two brook charr competitors in the littoral habitat, were introduced by bait fishers in many of these lakes over the last century. The objectives of this study were to test that (i) resource polymorphism is common in these brook charr populations, (ii) the presence of introduced species will decrease the phenotypic divergence between the two brook charr ecotypes, and (iii) the ecological release from creek chub and white sucker will increase population and/or individual niche widths in brook charr. The study was based on 27 lakes and five indicators of resource use (stomach content, liver δ13C, muscle astaxanthin concentration, pyloric caecum length, and gill raker length). Our results indicate that within-lake differences in resource use by both ecotypes are common and stable through time. When facing interspecific competition, both littoral and pelagic brook charr incorporated more pelagic prey into their diet but maintained the amplitude of their differences in resource use, which contradicts our second prediction. Finally, we did not find any significant effect of introduced species on population and individual niche widths of brook charr. We suggest that the difference in feeding mode among distantly related competitors could prevent the complete exclusion of a species from a given niche and explain the lack of response to ecological release.

Keywords: ecological release, phenotypic plasticity, resource partitioning, resource specialization, human-mediated introductions.

Introduction

Mechanisms underlying species diversification were first addressed in *On the Origin of Species* (Darwin 1859), but this question is still one the most challenging in evolutionary ecology (Pfennig et al. 2010; Wellborn and Langerhans 2015; Schneider and Meyer 2017; Skúlason et al. 2019). Ecological release following ecological opportunity would be the main factor promoting increases in phenotypic variance and, eventually, adaptive diversification (Yoder et al. 2010; Wellborn and Langerhans 2015). Ecological opportunity occurs when a resource becomes available through a decrease in interspecific competition or predation and when another species is capable of colonizing the vacant niche (i.e., ecological release) through phenotypic plasticity and intraspecific competition (Schluter 2000; Pfennig et al. 2010; Yoder et al. 2010; Des Roches et al. 2011). Its consequence should be population niche expansion via increased individual niche widths and/or increased among-individual variation (Bolnick et al. 2007; Bolnick et al. 2010).

Resource polymorphism, i.e., when discrete phenotypes of the same population show differential niche use, is a good example of the diversifying effect of intraspecific competition and may represent an important intermediate stage in sympatric speciation (Smith and Skúlason 1996; Skúlason et al. 2019; Schluter 2000). Common examples of this phenomenon are fish species found in depauperate post-glacial lakes of the northern hemisphere. These lakes almost always include coexisting benthic and pelagic ecotypes that are better adapted to feeding on bottom organisms or zooplankton, respectively (Robinson and Wilson 1994; Smith and Skúlason 1996). A first factor promoting resource polymorphism would be the exploitation of a new or unexploited resource to reduce intraspecific competition through phenotypic plasticity and divergent selection (selection against intermediates) (Smith and Skúlason 1996; Svanbäck and Bolnick 2007; Martin and Pfennig 2010; Araújo et al. 2011; Skúlason et al. 2019). A second factor would be a release from interspecific competition (Smith and Skúlason 1996; Bolnick et al. 2007; Skúlason et al. 2019), referred to as ecological opportunity.

In the context of resource polymorphism, one can predict an increase in phenotypic divergence between/among ecotypes when competitors are absent or, conversely, a decrease in phenotypic divergence when competitors are introduced (e.g., human-mediated introductions and species colonization). For example, pumpkinseed sunfish (*Lepomis gibbosus*) exhibit resource polymorphism in some Adirondack lakes (New York State, USA), with a limnetic form that inhabits open-water environments along with a littoral form found in the shallow littoral zone (Robinson et al. 1993). In a study on 11 lakes of the same area, Robinson et al. (2000) found that pumpkinseed fish were present in both habitats and were ecologically (diet and parasite load) and morphologically divergent in nine lakes where they were allopatric while they were restricted to the littoral zone in sympatry with bluegill sunfish (*L. macrochirus*). Similarly, in a survey on resource polymorphism in freshwater fishes of northern North America, Robinson and Schluter (2000) found that the distribution of phenotypes within each population was not always bimodal (i.e., littoral and limnetic ecotypes), but instances of polymorphism were found to be inversely related to total fish species diversity.

The ecological opportunity hypothesis has been supported by many empirical studies involving closely related species (Schluter 2000; Pfennig et al. 2010; Yoder et al. 2010; Wellborn and Langerhans 2015; Schneider and Meyer 2017; Skúlason et al. 2019). It is assumed that competition between closely related species will be stronger than between distantly related ones because the former are functionally similar, being close phylogenetically, and their niche will overlap almost completely. However, Schluter (2000) argued that the demonstration of character displacement between distant taxa should be a strong indication that members of one lineage constrain the phenotypic evolution in members of another lineage. The number of cases involving members of different taxa (genera or higher) is still small (Schluter 2000 and above-cited reviews), and evidence that competition is the mechanism of character displacement is rarely found (Schluter 2000). Since there have been very few cases studied involving distantly related taxa where one lineage affects the direction of habitat use of another lineage (Schluter 2000), investigating the interactions of these factors in natural systems is important for understanding the mechanisms underlying ecological release.

Brook charr exhibit a subtle resource polymorphism in some Canadian Shield lakes (Bourke et al. 1997; Dynes et al. 1999; Proulx and Magnan 2004; Pépino et al. 2018). The littoral ecotype has a deeper body and longer pectoral fins, is found in shallow water (0-3 m), and feeds mainly on zoobenthos. The pelagic ecotype has a more streamlined body shape with shorter pectoral fins, is found in deeper waters (4-6 m), and feeds mostly on zooplankton. Morphological differences between ecotypes seem to be determined by both genetic and environmental factors (Dynes et al. 1999; Proulx and Magnan 2004; Pépino et al. 2018). It has also been observed that morphological differentiation associated with resource polymorphism is functionally related to the swimming performance and energetics of the two ecotypes (Rouleau et al. 2010).

During the last century, bait fishers introduced creek chub (*Semotilus atromaculatus*) and white sucker (*Catostomus commersonii*) to many Canadian Shields lakes. These species compete for food with brook charr in the littoral zone (Magnan 1988; Tremblay and Magnan 1991; Lacasse and Magnan 1992). In these lakes, the level of interspecific competition varies from none (allopatric brook charr lakes), to intermediate (brook charr with creek chub lakes), to high (brook charr with creek chub and white sucker lakes) (Magnan 1988; Bourke et al. 1999; Magnan et al. 2005). Lakes with only white sucker are scarce, probably because both white sucker and creek chub were commonly used by bait fishers when this practice was popular (although it was never authorized). This system thus represents a good model to investigate the effects of interspecific competition and ecological release (allopatric vs. sympatric brook charr populations) on the expression of resource polymorphism. Wellborn and Langerhans (2015) also emphasized that strong and widespread environmental impacts of humans, like the introduction of novel competitors, could foster high levels of diversification in organisms, but that it may also modify factors that influence population responses to ecological opportunity. Given the prevalence of species introductions in many ecosystems around the world (e.g., Pysek et al. 2010), it is relevant to address their implications on the process of species diversification. We predict (i) that resource polymorphism will be common in brook charr populations of Canadian Shield lakes, (ii) that the presence of creek chub and white sucker will decrease the phenotypic divergence between littoral and pelagic brook charr because

introduced species will restrict individuals in their use of littoral zone resources, and (iii) that ecological release from creek chub and white sucker will increase population and/or individual niche widths in brook charr (e.g., Bolnick et al. 2010). To this end, we sampled brook charr from 27 Canadian Shield lakes along the gradient of interspecific competition described above to study the impacts of species introduction (i.e., the reverse of ecological release) on the expression of resource polymorphism. We used five indicators of resource use (i.e., stomach content, stable isotope, carotenoid pigment, pyloric caecum length, and gill raker length) that exhibit different degrees of temporal integration (i.e., from days to months to seasons) to characterize resource specialization. Although some of these indicators were used independently in fewer lakes of the same system (see Materials and Methods section), this approach allowed us to address the temporal stability of resource specialization at a larger spatial scale.

Materials and methods

Study area and fish sampling

The study was conducted in the Mastigouche (46° 40′ N, 73° 30′ W) and Saint-Maurice (47° 05′ N, 73° 15′ W) wildlife reserves and in La Mauricie National Park (46°45′ N, 73°08′ W), Québec, Canada, from June to August 2012, 2013, and 2014. We sampled 10 lakes containing only brook charr (BC), eight with brook charr and creek chub (BC+CC), and nine with brook charr, creek chub, and white sucker (BC+CC+WS) (Table 1). With one exception (Lake Dorval), CC always occur with WS. Other species were found in BC+CC and BC+CC+WS lakes, but the main competitors of brook charr in this system are creek chub and white sucker (Magnan 1988; Magnan et al. 2005). Each lake was sampled only once except for lakes Baie des Onze Îles, Baie Verte, Baie Cobb, and Maréchal, which were sampled twice in 2012 because unusually warm water temperatures limited the capture of brook charr in the littoral zone (see details below). Fish were caught in both the littoral (< 3 m) and pelagic (> 4 m) zones of lakes. The depth of 3 m represents the limit of the littoral zone in this system, as defined by Wetzel and Likens (2000), i.e., "the zone containing a substantial number of microhabitats

associated with surfaces of submersed macrophytes, with particulate detritus, and with the sediments." We arbitrarily considered the beginning of the pelagic zone at a depth > 4 m to prevent any overlap with the littoral zone. The mean depths (± SD) of the trap nets were 2.1 ± 0.8 m and 5.2 ± 1.0 m in the littoral and pelagic zones, respectively.

We used two trap nets per zone (Alaska type; opening 1.0 m × 1.8 m, equipped with two 1 m × 15 m wings; Fipec Industries, Gaspé, Québec, Canada). The traps were located randomly in each zone and fished for four (2013, 2014) or five (2012) days from 18:00 to 06:00 to capture brook charr at the time they were most active (Bourke et al. 1996). Based on previous studies in this system, this protocol was appropriate to maximize the chances of capturing littoral brook charr in the littoral zone and pelagic brook charr in the pelagic zone because littoral individuals return to the metalimnion during the day for thermoregulation needs (Goyer et al. 2014). The mean depth (± SD) of the trap nets were 2.1 ± 0.8 m and 5.2 ± 1.0 m in the littoral and pelagic zones, respectively. We sampled a maximum of 60 brook charr in each zone, with a mean of 24 fish per lake × zone × sex. This sample size was based on our knowledge of this system to get representative data, especially for diet analyses. These lakes are subject to sport fishing, and their exploitation is carefully controlled by governmental agencies. Fish sampled for this study were subtracted from the annual fishing quota of each lake so that no additional fish were captured above the maximum sustained yield. Each charr was measured (fork length), weighed, and sexed after being euthanatized with an excess of eugenol (clove oil, 200 mg/L; Pounder et al. 2018). Additional biological samples were taken from these individuals (see details below).

Stomach content analysis

The digestive tracts of brook charr were transferred to a 4% formalin solution immediately after capture. The stomach contents (i.e., the part of the digestive tract situated between the oesophagus and the pyloric valve) of 2583 brook charr from 27 lakes were analyzed under a dissecting microscope. We removed 958 empty stomachs from this dataset, resulting in a final sample of 1625 individuals for diet analyses. Prey were sorted into

nine categories based on their functional attributes (zooplankton, macroinvertebrates, amphipods, dipteran pupa, swimming insects, terrestrial insects, prey fish, leeches, and others; Lacasse and Magnan 1992). The weight (± 1.0 mg) of each category was noted after drying for 48 h at 60°C. The mean percent weight of each prey category (Hyslop 1980) was used to represent the diet of littoral and pelagic individuals while the proportion of pelagic prey (zooplankton and dipteran pupae) in the stomach of each individual (pelagic prey weight/stomach content weight) was used as the dependent variable in the mixed models (see Statistical analyses section). The proportion of pelagic prey ranged from 0 (i.e., only littoral prey in the stomach) to 1 (i.e., only pelagic prey in the stomach). We excluded terrestrial insects (e.g., butterfly) and the category "others" from this proportion because they can be found in both zones. We also excluded prey fish (mostly northern redbelly dace) and leeches from the analyses because brook charr seem to feed on them once inside the trap net (V. Rainville, personal observation) and thus they represent a bias induced by the capture method rather than a natural prey. These are not common brook charr prey according to gillnet captures (Magnan 1988; East and Magnan 1991; Lacasse and Magnan 1992).

Stomach content analysis is among the most common indicators of resource use because it provides direct and detailed information on short-term (24 h-48 h) prey consumption. Because prey categories of the littoral and pelagic zones are spatially isolated, the short-term nature of stomach content analysis also means that we are less likely to incorporate prey categories of both zones. Therefore, although stomach content provides useful information on resource use, it needs to be complemented with longer-term indicators to infer specialization in feeding habits.

Stable isotope analysis

Stable isotope analysis of carbon can be used as a space- and time-integrated indicator of a consumer's assimilated carbon sources (Glaz et al. 2012; Layman et al. 2012). The carbon signature, expressed in delta notation ($\delta^{13}C$), is defined as the deviation from an international standard material (Vienna Pee Dee Belemite) in parts per thousand (‰).

In post-glacial lakes, the $\delta^{13}C$ of species associated with the pelagic habitat tends to be more negative (i.e., ^{13}C-depleted) than species associated with the littoral habitat (Glaz et al. 2012). Therefore, the carbon signature of a consumer can be used to estimate the relative contribution of both littoral and pelagic resources at an individual level. Furthermore, different tissues can provide information on the assimilated prey over different time scales (Layman et al. 2012). For instance, in arctic charr *Salvelinus alpinus*, liver tissue has a response time of about a month while muscle tissue (depending on growth) showed a response time of about four to five months (Perga and Gerdeaux 2005).

The isotopic signature of primary producers is known to be highly variable. Therefore, we used primary consumers as a baseline to obtain good approximations of long-term carbon sources (Vander Zanden and Rasmussen 1999). For each lake, we used zooplankton collected with Wisconsin net tows in the pelagic zone and macroinvertebrates from different orders (i.e., Amphipoda, Ephemeroptera, Trichoptera, Anisoptera, and Zygoptera) captured in the littoral zone using a dip net. All samples were frozen on dry ice (-78.5°C) immediately after collection and subsequently stored at -20°C. Samples were thawed at room temperature, sorted, and cleaned under a dissecting microscope to remove periphyton, detritus, and phytoplankton. Each sample was then dried for 48 h at 60°C and ground into a fine powder using a mortar and pestle (Glaz et al. 2012).

The livers from brook charr captured in both the littoral and pelagic zones were dissected immediately after their capture and euthanasia, and they were processed like the primary consumers. A subsample of 840 livers from 22 lakes was used in statistical analyses (Table 1). Each 1 mg sample (primary consumers and brook charr liver) was packed into a tin capsule (Elemental Microanalysis 8 × 5 mm) and analyzed with a mass spectrometer (Thermo Scientific model Delta V Plus Isotope Ration MS). We used USGS40 (L-glutamic acid: $\delta^{13}C$ mean ± SD = -26.389 ± 0.042) from the International Atomic Energy Agency (IAEA) as the $\delta^{13}C$ standard. $\delta^{13}C$ values from brook charr liver were corrected for lipid bias using the C:N ratio as in Post et al. (2007).

Carotenoid analysis

Zooplankton accumulate high concentrations of carotenoid pigments (Green 1957; Brüsin et al. 2016). Like all vertebrates, fishes cannot synthesize carotenoids and thus must acquire them through their food intake (Goodwin 1952). In salmonids, the reddish colouration of the flesh is due to carotenoid pigments, especially astaxanthin, the primary pigment found in wild individuals (Meyers 1994). Carotenoid pigments could serve as a relatively short-term indicator (~two months; Saito and Regier 1971) of pelagic resource use in freshwater fish.

Carotenoid concentrations in brook charr muscle were determined using two methods. We first estimated the carotenoid concentrations in 100 brook charr captured in the littoral and pelagic zones of two BC and three BC+CC+WS lakes (five males and five females per zone per lake; Table 1) using high performance liquid chromatography (HPLC). Approximately 5 g of muscle were sampled between the dorsal and adipose fins and the lateral line. To prevent pigment degradation from oxygen, light, and heat (Choubert et al. 2011), the muscle was wrapped in skin sampled from the same fish, vacuum packed in a sterile, chemically inert polyethylene bag (2 oz. Whirl-Pak from Nasco) immediately after capture, and frozen on dry ice (-78.5°C) before being transferred to a freezer in our laboratory (-20°C). The two main pigments found in salmonids are astaxanthin and canthaxanthin (Meyers 1994). However, only astaxanthin was estimated in our study because canthaxanthin was almost always below the detection threshold of our chromatograph (Waters, Detector UV-Visible 2-Channel Model 2487). A sample of about 0.6 g of frozen muscle was finely crushed and transferred into a 20 ml amber vial that was then filled with acetone. The vial was agitated with a microplate shaker (Brinkmann; Orbimix 1010) for 1 h and then stored at 4°C. About 24 h later, samples were filtered and evaporated in a roto-evaporator (Brinkmann; rotavapor R110) to concentrate pigments in 5-10 ml of acetone (Strati et al. 2012). Muscle particles were pure white after the filtration, indicating that most pigments were extracted. As recommended by the column manufacturer, we used two eluents: methanol / methyl tert-butyl ether / demineralized water (81/15/4) (eluent A) and methanol / methyl tert-butyl ether / demineralized water (6/90/4) (eluent B). Pigments were injected into a binary HPLC pump (Waters,

Model 1525) and eluted using a linear gradient from 100% of eluent A to 100% of eluent B for 90 min at a flow rate of 1.0 ml/min. Pigments were detected at 450 nm with a Waters Detector UV-Visible 2-Channel Model 2487. Analytical grade acetone was used for sample extraction (Fisher Scientific Ottawa, Canada). HPLC grade methyl tert-butyl and methanol were obtained from Sigma-Aldrich Canada Co (Oakville, ON, Canada) and Fisher Scientific (Ottawa, ON, Canada), respectively. Standard dilutions of 0.5-7 μg/ml of acetone were prepared for the calibration curve; they were stored at -20°C and were stable throughout the study. Due to problems in pigment separation, two samples (out of 100) were not used.

Since the HPLC method is time-consuming and expensive, we also estimated the muscle colouration of 1533 brook charr from 17 lakes (Table 1) with the Roche SalmoFan© lineal card (see example from Lake Maréchal, Fig. 1). This chart is widely used for the visual assessment of fish fillets for quality control in the salmonid industry. Scores of the Roche SalmoFan© lineal card are associated with the carotenoid concentration (especially astaxanthin) and range from 20 (pale pink) to 34 (dark red). Readings were done in the field on adult charr muscle (~ 20 mm × 30 mm) sampled immediately after capture and euthanasia. Muscles were placed between a white and a transparent Plexiglas plate separated by 0.7 cm so that muscle thicknesses were comparable among samples. Because the Roche SalmoFan© lineal card is designed for the salmonid industry, it did not cover the complete range of colours for individuals in our study lakes (I. Lussier, personal observation). We thus assigned a value 18 for muscles scoring below 20 on the chart (30 littoral and 17 pelagic individuals). No flesh colouration was estimated above the maximum value of the chart. For each lake, SalmoFan© lineal card scoring was done by the same observer, and two observers scored the 17 studied lakes (considered as a fixed effect in the statistical analyses; see below).

Pyloric caecum and gill raker analyses

Pyloric caeca are known to be important sites of nutrient absorption and production of digestive enzymes in salmonids (Buddington and Diamond 1986; Krogdahl et al. 1999).

Magnan and Stevens (1993) observed that brook charr caeca were 17.4% longer in lakes containing white sucker, where individuals fed more on zooplankton than on benthic prey. Differences in pyloric caecum length were also related to resource specialization in two ecotypes (littoral and limnetic) of arctic charr (Knudsen et al. 2008) and perch (Olsson et al. 2007). Since creek chub and white sucker were introduced to these lakes during the last century, it would be plausible to observe an increase in the length of brook charr pyloric caeca in lakes with interspecific competition due to a phenotypic response to their increased feeding on zooplankton. The pyloric caeca of brook charr were transferred to a 4% formalin solution immediately after capture and euthanasia. We measured the length of the last pyloric caecum, located at the end of the anterior intestine (Magnan and Stevens 1993). These measurements were made to the nearest 0.1 mm using an ocular micrometer mounted on a 10x dissection microscope. We randomly selected 15 individuals from each zone of all lakes sampled in 2014 (n = 320; Table 1).

Finally, long and dense gill rakers may improve particle retention of small prey like zooplankton, and there is experimental evidence that these characteristics are adaptations for feeding more efficiently on planktonic prey (Robinson et al. 1993; Roesch et al. 2013). Magnan (1988) observed that brook charr gill raker lengths were significantly longer in lakes containing creek chub (7.2%) and white sucker (19.7%), where individuals fed more on zooplankton than zoobenthos. Differences in gill raker length, separation, and/or number between limnetic and benthic ecotypes have been observed in other fishes, such as *Coregonus* spp. (Amundsen 1988), arctic charr (*Salveninus alpinius*) (Malmquist 1992), rainbow smelt (*Osmerus mordax*) (Taylor and Bentzen 1993), and bluegill sunfish (*Lepomis macrochirus*) (Robinson et al. 1993). The gill arches of brook charr were transferred to a 4% formalin solution immediately after capture and euthanasia. We measured the length of the longest gill raker at the bend in the left gill arch (Magnan 1988) to the nearest 0.1 mm using an ocular micrometer mounted on a 10x dissection microscope. We randomly selected 15 individuals from each zone of all lakes sampled in 2014 and five individuals from each zone of all lakes sampled in 2013 (n = 390; Table 1).

Niche breadth components

The total niche width (TNW) of a population represents the full spectrum of prey species consumed in a given population and is the sum of two components: the within-individual component (WIC), which is the dietary variation of a typical individual, and the between-individual component (BIC), representing the dietary variation among individuals (Roughgarden 1972). We estimated these components from the diet of brook charr in BC, BC+CC, and BC+CC+WS lakes using the R package RInSp (Zaccarelli et al. 2013). Two lakes, Lézard ($n = 8$) and Dorval ($n = 2$), were excluded from the statistical analyses due to their low sample sizes (Table 1).

Statistical analyses

Due to unequal sample sizes among the five indicators, we analyzed each response variable separately using a mixed-effects modelling approach (detailed below). Considering that samples were not taken on the same individuals for all indicators, the use a global model would have reduced the sample size to 165 instead of 2583 brook charr for the stomach contents, 840 for $\delta^{13}C$, 1533 for carotenoids (SalmoFan lineal Card), 320 for pyloric caeca, and 390 for gill rakers., The within-lake Pearson correlation among the five indicators (computed when sample size was > 20) averaged 0.14 and ranged from -0.33 (between caecum length and gill raker length) to 0.53 (between carotenoid concentration and caecum length). All indicator variables were size adjusted before calculating the Pearson correlation, which was particularly important for caecum length and gill raker length. We are thus confident that the different models described below are not redundant and bring complementary information concerning the expression of resource polymorphism and integrate different temporal scales.

We used a mixed-effects modelling approach to account for the hierarchical structure of the data, with individual nested in lake and sampling year. We built a sequence of increasingly complex models including various combinations of fixed-effect variables (Table 2). These models included two variables of interest as predictors: the zone of capture (Z) as a proxy of phenotypic divergence between ecotypes (effect coding; littoral

and pelagic as -1 and 1, respectively) and the fish community (C) as a proxy of interspecific competition for littoral resources (dummy variable with BC for brook charr as the reference category, BC+CC for creek chub, and BC+CC+WS for white sucker lakes). Although the relative abundance (catch per unit of effort) of competitors shows some variation among lakes, the relationship between the level of interspecific competition and the occurrence of WS and/or CC is stable in this system (Magnan 1988; Bourke et al. 1999; Magnan et al. 2005). In this context, the present study investigated the mode of competition instead of any density-dependent effect of interspecific competition. Because brook charr display sexual dimorphism, we included the sex (S; effect coding; female and male as -1 and 1, respectively) in models where we had a priori ecological hypotheses (Table 2). All models included standardized (i.e., $\mu = 0$, $\sigma = 1$) and log-transformed ($\log_e x$) fish length to take into account possible ontogenetic resource shifts in our samples, even though the size distribution of individuals was relatively narrow (mean ± SD: 21.5 ± 5.3 cm). We also included the standardized day of year (D) in all models to account for resource seasonality. One model (model 11; Table 2) includes the interaction between day of year and fish community because brook charr experience higher competition from creek chub and white sucker in the littoral zone as the summer progresses (Tremblay and Magnan 1991; Lacasse and Magnan 1992) due to the seasonal decline in the zoobenthic community, which generally occurs in temperate lakes through summer (Mittelbach 1981; Persson 1987). Because stomach contents and stable isotopes are expressed as proportions (see section below for isotopic mixing models), we used generalized linear mixed models (R package glmmTMB, v0.2.0.9; Magnusson et al. 2017) with beta distributions and a logit link function.

We used linear mixed models (function lmer of the R package lme4; Bates et al. 2014) for caecum and gill raker lengths, carotenoid concentration, and muscle colour. Model comparison was based on Akaike Information Criterion adjusted for sample size (AICc; Burnham and Anderson 2002). Models were ranked using ΔAICc, i.e., the difference in AICc between a candidate model and the model with the lowest (best) AICc. We selected the most parsimonious model within two ΔAICc units of the best model, but we also discuss parsimonious models within four ΔAICc units.

The relative contributions of pelagic and littoral prey on the $\delta^{13}C$ of brook charr livers were estimated using stable isotope analysis in R (SIAR, v4.2) mixing models (Parnell and Jackson 2013). Stable isotope mixing models allow the incorporation of natural variability in the $\delta^{13}C$ baseline of organisms as well as the trophic enrichment factors (Parnell et al. 2010). We assumed a trophic enrichment of 0.39 ± 1.3‰ for ^{13}C in our system (Post 2002). These mixing models also provide probability distributions of the relative contribution of possible sources (i.e., littoral vs. pelagic) at an individual level that can then be used in further analysis (Parnell et al. 2010; Layman et al. 2012). To account for the propagation of uncertainty around the estimation of littoral vs. pelagic contribution for each individual, we randomly selected one value for each individual from the SIAR mixing model and used it in the mixed modelling approach (as described above). We ran 1000 iterations of this last step to assess (i) if one of the competing mixed models systematically stands out as the best model and (ii) if the estimates of this best model are reliable across the 1000 different input datasets.

We used ANOVAs to test the null hypotheses of no differences in TNW and WIC of brook charr among BC, BC+CC, and BC+CC+WS lakes. TNW and WIC were log-transformed prior to analyses to fulfill the condition of normality. Finally, we assessed the relationship between carotenoid concentrations measured with HPLC and muscle colour estimated with the Roche SalmoFan© using multiple linear regression models with log-transformed astaxanthin concentration as the response variable and the SalmoFan© score and observer as predictors.

Results

The most frequent and abundant prey categories in brook charr stomach contents were macroinvertebrates, zooplankton, and terrestrial insects (Fig. 2). The best model candidate predicting resource use from stomach contents includes the zone of capture and fish community as well as the interaction between day of the year and community (Table 2, model 11). This model predicts that brook charr captured in the pelagic zone consume 10% more pelagic prey than those captured in the littoral zone (Fig. 3a). It is noteworthy

that within any given fish community, some of the charr fed almost exclusively on benthic organisms ("benthic specialists"; mean percent weight of pelagic prey < 10%), others fed almost exclusively on pelagic prey ("pelagic specialists"; mean percent weight of pelagic prey >90%), and a very low proportion were "generalist feeders" (i.e., mean percent weight of benthic prey between 10 and 90%; Fig. 4). The model also predicts that the proportion of pelagic prey in brook charr stomachs increases from 38% in BC lakes to 49% in BC+CC lakes and to 55% in BC+CC+WS lakes (Fig. 3a). In the same way, the proportion of benthic specialists decreased from BC lakes to BC+WS lakes and to BC+CC+WS lakes and vice versa for the pelagic specialists (Fig. 4). While day of the year had no impact on pelagic prey consumption in BC lakes, this proportion increased with day of year in both BC+CC and BC+CC+WS lakes (Table 3).

The model predicting resource use from $\delta^{13}C$ with only the zone of capture was selected as the best model 75.1% of time in the process of iterations (Table 2, model 2). This model predicts that the relative pelagic contribution to the liver ranged from 0.207 in brook charr captured in the littoral zone to 0.273 in brook charr captured in the pelagic zone, corresponding to a 31.7% increase (Fig. 3b). Model estimates were stable across 1000 iterations (Fig. 5). In most cases, the mixing models successfully estimated the relative contribution of pelagic resources (narrow distributions). However, the individual contribution of pelagic resources to liver $\delta^{13}C$ estimated with the SIAR mixing models revealed high inter-individual variability in the estimated resource use in some lakes (Figs. 6-8).

The best model predicting astaxanthin concentration in muscle includes the zone of capture and fish community as well as the interaction between day of year and fish community (Table 2, model 11). The model predicts that individuals captured in the pelagic zone will have astaxanthin concentrations 41% higher than fish captured in the littoral zone (Fig. 3c). The model also predicts that individuals from BC+CC+WS lakes will have astaxanthin concentrations 57% higher compared to individuals from BC lakes. Finally, the astaxanthin concentration will increase with day of the year in BC+CC+WS lakes but decrease as summer progresses in BC lakes (Table 3). Other parsimonious

models also fall within the four ΔAICc threshold (Table 2, models 2 and 9). These models also include the zone of capture, and model 9 includes fish community. Two of the models predicting brook charr muscle colour from the Roche SalmoFan© lineal card fall under the two ΔAICc threshold. The first and most parsimonious model only includes the zone of capture (Table 2, model 2). This model predicts that muscle colour (a proxy for astaxanthin) of brook charr captured in the pelagic zone will be 0.3 SalmoFan© units higher compared to individuals captured in the littoral zone (Fig. 3d). The second model includes the zone of capture and fish community (Table 2, model 9); this model predicts a similar effect of the zone of capture and an additional effect of fish community: the muscle colour should increase by 1.3 SalmoFan© units from BC to BC+CC and by 3.0 SalmoFan© units from BC to BC+CC+WS lakes (Fig. 3d). The linear regression between astaxanthin concentration from HPLC and muscle colour from the SalmoFan© card explained 36% of the variation ($p < 0.001$; Table 4; Fig. 9). We also tested the interaction term between SalmoFan© scores and observers, but this interaction was not statistically significant ($p = 0.19$).

The best model predicting caecum length includes both zone of capture and fish community (Table 2, model 9). The model predicts that the caeca of individuals captured in the pelagic zone will be 6% longer compared to those captured in the littoral zone (Fig. 3e gives the order of magnitude of the effect size in mm). The model also predicts an increase of 9% in caecum length from BC to BC+CC and 31% from BC to BC+CC+WS lakes (Fig. 3e). Another model that only included fish community falls within the four ΔAICc threshold; it predicts similar effects for the fish community (Fig. 3e).

The best model predicting gill raker length includes only the fish community (Table 2, model 4). This model predicts an increase of 8% in gill raker length from BC to BC+CC and 10.5% from BC to BC+CC+WS lakes (Fig. 3f gives the order of magnitude of the effect size in mm). For comparative purposes with other indicators, we also included the effect size (i.e., the amplitude of differences predicted by the models) of the zone of capture in this figure as estimated by model 9 (Table 2). Despite the trend for longer gill

raker length in pelagic compared to littoral brook charr (i.e., 0.03 mm longer for pelagic individuals), no significant effect of the ecotype was detected in our mixed models.

The TNW and WIC varied more than two orders of magnitude among lakes and exhibited large overlaps among fish communities (Fig. 10). The TNW and WIC were not significantly different among the fish communities (Fig. 11; TNW: $F_{22,2\ (0,05)} = 0.296$, $p = 0.746$; WIC: $F_{22,2\ (0,05)} = 0.367$, $p = 0.697$).

Discussion

The results showed that resource specialization of littoral and pelagic brook charr is common in all study lakes (i.e., pelagic brook charr consumed more planktonic prey than did littoral brook charr in all fish communities) and that within-lake differences in resource use by both ecotypes are stable through time (i.e., even though all indicators have different integration time scales, they are all consistent with our prediction, with the exception of gill raker length which exhibit little differences between littoral and pelagic brook charr), supporting our first prediction.

The results also showed a major impact of interspecific competition (among-lake variation) on resource use. When facing competition by creek chub and/or white sucker, both littoral and pelagic brook charr incorporated more pelagic prey into their diets but maintained their differences in resource use (within-lake variation). The consumption of pelagic prey by sympatric brook charr also increased as the summer progressed, likely related to the progressive summer decline in zoobenthos that generally occurs in temperate lakes (Mittelbach 1981; Persson 1987). Models including the interaction between ecotype and fish community were never retained as the best models to explain variations in the five indicators of resource use, indicating that the magnitude of the difference in resource use between littoral and pelagic ecotypes is similar along the gradient of interspecific competition. This contradicts our second prediction – that interspecific competition will decrease the phenotypic divergence between littoral and pelagic brook charr by restricting individuals to resources in the open-water zone.

Finally, while some individuals clearly specialized on either littoral or pelagic resources, most individuals used both resources over a longer temporal scale (based on stable isotopes) than what is revealed by stomach contents. The fact that fish community was not retained as a significant factor in the stable isotope analysis suggests that the proportion of benthic and pelagic prey used by individuals of both ecotypes is comparable along the gradient of interspecific competition over weeks. These results could reflect a flexibility in resource specialization of individuals from both ecotypes that is related either to an opportunistic use of overabundant resources at given periods of the year (e.g., mayfly emergence) or to the progressive summer decline of the zoobenthic communities mentioned above. However, despite this flexibility in resource specialization by individuals from both ecotypes, pelagic brook charr consumed more planktonic prey than did littoral brook charr in all fish communities, and this difference was stable through time. Such a replicated resource polymorphism in brook charr of the Canadian Shield might preclude future adaptive radiation in these lakes. Wellborn and Langerhans (2015) suggested that cases of replicate adaptive radiation in the wild point to strong determinism in the action of ecological opportunity. These patterns of adaptive radiation demonstrate that similar habitats, comprising similar arrays of niches, yield similar patterns of ecological diversification (i.e., parallel evolution).

Yoder et al. (2010) outlined three main characteristics of ecological release: populations should exhibit (i) density compensation resulting from decreased interspecific competition in species-poor habitats, (ii) broadened habitat or resource use, probably in concert with density compensation, and (iii) increased trait variation. In our system, competition by creek chub and white sucker reduced the relative abundance and biomass of brook charr by 30% to 70% (Magnan 1988; Lachance and Magnan 1990; Magnan et al. 2005), which is consistent with ecological release. Overall, the effect of interspecific competition was stronger than the differences between ecotypes (i.e., zone of capture effect), with both littoral and pelagic brook charr incorporating more pelagic prey in BC+CC and BC+CC+WS lakes than in BC lakes. However, both ecotypes maintained the magnitude of their differences in resource use along the gradient of interspecific competition (no interaction between ecotype and fish community in the best models), indicating no

resource broadening in the absence of interspecific competition. These results are thus contrary to the last two predictions of ecological release presented above.

Our third prediction was that the ecological release from creek chub and white sucker will increase population and/or individual niche widths in brook charr. Bolnick et al. (2010) presented three potential scenarios of ecological release. Release from competition can lead to (i) increased individual (WIC) and population (TNW) niche widths (parallel release), (ii) increased TNW but not WIC via increased among-individual variation (niche variation hypothesis), or (iii) increased WIC but not TNW: because expansion is offset by lower among-individual variation (individual release), the population niche width remains unchanged. The first two scenarios correspond to the classical view of ecological release (Bolnick et al. 2010). For the third scenario, Bolnick et al. (2010) mentioned that it could be counterintuitive to think that ecological release could reduce prey availability, but it has been suggested that this can arise if intraspecific competition increases disproportionately following release (e.g., Trewby et al. 2007). We did not find any significant difference in TNW and WIC between BC lakes (ecological release) and BC+CC or BC+CC+WS (competition from introduced species).

We suggest that the competitive release from creek chub and white sucker led to a fourth scenario in brook trout: no change in individual and population niche width following ecological release. This scenario could be explained by a combination of different mechanisms. First, the intraspecific competition in these depauperate oligotrophic lakes could be strong enough to constrain the individual niche width of each ecotype in their respective habitats (littoral and pelagic) following competitive release. Although populations in species-rich communities can exhibit high trait/niche variation (e.g., Mittelbach et al. 2007; Losos 2010), there is evidence that high trait variance tends to occur in depauperate environments and communities (Schluter 2000; Wellborn and Langerhans 2015). This is particularly true for fish inhabiting postglacial lakes of the Northern Hemisphere (Skúlason and Smith 1995). These lakes offer two discrete functional habitats – the littoral and pelagic zones – and studies of resource polymorphisms almost always include co-existing benthic and pelagic ecotypes

(Robinson and Wilson 1994; Skúlason and Smith 1995). Second, the feeding mode of brook charr (a particulate feeder) differs from that of white sucker (bottom suction feeder), and even though creek chub is a particulate feeder, it is gape-limited in prey size compared to brook charr. These differences could leave a portion of the littoral niche available for brook charr in lakes with introduced species, explaining why some individuals still use this habitat in sympatry (in contrast to ecological segregation between bluegill and pumpkinseed sunfish in Adirondack lakes; Robinson et al. 2000). Of course, the carrying capacity of the littoral zone for brook charr is lower in sympatric populations, explaining why their abundance and biomass is lower in these communities (Magnan 1988; Lachance and Magnan 1990; Magnan et al. 2005). These results suggest that the level of intraspecific competition among brook charr remains comparable in allopatric and sympatric populations, explaining why the two ecotypes are maintained in sympatry and why resource specialization remains of the same magnitude in both communities. Such a response following ecological release (i.e., no change in individual and population niche width) could represent one outcome in depauperate environments, especially when competitors are distantly related (i.e., the fourth scenario presented above).

Our results suggest that the effects of interaction between distantly related species could be quite different from those between closely related species due, for example, to higher asymmetry in their feeding modes, preventing the exclusion of one species from a niche it exploited in allopatry (i.e., in the situation of competitive release). This study also highlights that human-mediated introductions have the potential to alter the trophic niche of native species. We used the comparison of lakes with and without introduced species to test if ecological release from distantly related species affects population and individual niche widths in brook charr. This is of particular interest considering the human propensity to introduce exotic species in pristine ecosystems, thus decreasing ecological opportunity and changing the fate of species diversification. In this context, it would be relevant to continue to develop and test conceptual frameworks of ecological opportunity dealing with human-mediated introductions of distantly related species in future studies.

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References

Amundsen P-A (1988) Habitat and food segregation of two sympatric populations of whitefish (*Coregonus lavaretus* L. *s.l.*) in Stuorajavri, Northern Norway. Nordic J Freshw Res 64:67-73.

Araújo MS, Bolnick DI, Layman CA (2011) The ecological causes of individual specialisation. Ecol Lett 14:948-958. doi: 10.1111/j.1461-0248.2011.01662.x

Bates D, Maechler M, Bolker B, Walker S (2014) lme4: Linear mixed-effects models using Eigen and S4. R package version 1.1-21. http://CRAN.R-project.org/package=lme4

Bolnick DI, Svanbäck R, Araújo MS, Persson L (2007) Comparative support for the niche variation hypothesis that more generalized populations also are more heterogeneous. Proc Natl Acad Sci USA 104:10075-10079. doi: 10.1073/pnas.0703743104

Bolnick DI, Ingram T, Stutz WE, Snowberg LK, Lau OL, Paull JS (2010) Ecological release from interspecific competition leads to decoupled changes in population and individual niche width. Proc R Soc B Biol Sci 277:1789-1797. doi: 10.1098/rspb.2010.0018

Bourke P, Magnan P, Rodriguez MA (1996) Diel locomotor activity of brook charr, as determined by radiotelemetry. J Fish Biol 49:1174-1185. doi:10.1006/jfbi.1996.0245

Bourke P, Magnan P, Rodriguez MA (1997) Individual variations in habitat use and morphology in brook charr. J Fish Biol 51:783-794. doi: 10.1006/jfbi.1997.0481

Bourke P, Magnan P, Rodriguez MA (1999) Phenotypic responses of lacustrine brook charr in relation to the intensity of interspecific competition. Evol Ecol 13:19-31.

Brüsin M, Svensson PA, Hylander S (2016) Individual changes in zooplankton pigmentation in relation to ultraviolet radiation and predator cues. Limnol Oceanogr 61:1337-1344. doi: 10.1002/lno.10303

Buddington RK, Diamond JM (1986) Aristotle revisited: The function of pyloric caeca in fish. Proc Natl Acad Sci USA 83:8012-8014.

Burnham KP, Anderson DR (2002) Model selection and multimodel inference: A practical information-theoretic approach. Springer-Verlag, New York

Choubert G, Brisbarre F, Baccaunaud M (2011) Impact of dietary carotenoid and packaging during frozen storage on the quality of rainbow trout (*Oncorhynchus mykiss*) fed carotenoids. J Sci Food Agric 91:1075-1082. doi: 10.1002/jsfa.4286

Darwin C (1859) On the origin of species by means of natural selection. London: John Murray.

Des Roches S, Robertson JM, Harmon LJ, Rosenblum EB (2011) Ecological release in White Sands lizards. Ecol Evol 1:571-578. doi: 10.1002/ece3.50

Dynes J, Magnan P, Bernatchez L, Rodriguez MA (1999) Genetic and morphological variation between two forms of lacustrine brook charr. J Fish Biol 54:955-972.

East P, Magnan P (1991) Some factors regulating piscivory of Brook Trout, *Salvelinus fontinalis*, in lakes of the Laurentian Shield. Can J Fish Aquat Sci 48:1735-1743. doi: 10.1139/f91-205

Glaz P, Sirois P, Nozais C (2012) Determination of food sources for benthic invertebrates and brook trout *Salvelinus fontinalis* in Canadian Boreal Shield lakes using stable isotope analysis. Aquat Biol 17:107-117. doi: 10.3354/ab00465

Goodwin T (1952) The comparative biochemistry of carotenoids. Chapman & Hall LTD., London.

Goyer K, Bertolo A, Pépino M, Magnan P (2014) Effects of lake warming on behavioural thermoregulatory tactics in a cold-water stenothermic fish. PLoS One 9:e92514. doi: 10.1371/journal.pone.0092514

Green J (1957) Carotenoids in Daphnia. Proc R Soc B Biol Sci 147: 392-401. doi: 10.1098/rspb.1957.0058

Hyslop EJ (1980) Stomach contents analysis - a review of methods and their application. J. Fish Biol. 17: 411-429.

Knudsen R, Amundsen PA, Jobling M, Klemetsen A (2008) Differences in pyloric caeca morphology between Arctic charr *Salvelinus alpinus* ecotypes: Adaptation to trophic specialization or parasite-induced phenotypic modifications? J Fish Biol 73:275-287. doi: 10.1111/j.1095-8649.2008.01934.x

Krogdahl Å, Nordrum S, Sørensen M, Brudeseth L, Røsjø C (1999) Effects of diet composition on apparent nutrient absorption along the intestinal tract and of subsequent fasting on mucosal disaccharidase activities and plasma nutrient concentration in Atlantic salmon *Salmo salar* L. Aquac Nutr 5:121-133. doi: 10.1046/j.1365-2095.1999.00095.x

Lacasse S, Magnan P (1992) Biotic and abiotic determinants of the diet of brook trout, *Salvelinus fontinalis*, in lakes of the Laurentian Shield. Can J Fish Aquat Sci 49:1001-1009.

Lachance S, Magnan P (1990) Performance of domestic, hybrid, and wild strains of brook trout, *Salvelinus fontinalis*, after stocking: the impact of intra- and interspecific competition. Can J Fish Aquat Sci 47:2278-2284. doi: doi.org/10.1139/f90-253

Layman CA, Araujo MS, Boucek R, Hammerschlag-Peyer CM, Harrison E, Jud ZR, Matich P, Rosenblatt AE, Vaudo JJ, Yeager LA, Post DM, Bearhop S (2012) Applying stable isotopes to examine food-web structure: An overview of analytical tools. Biol Rev 87:545-562. doi: 10.1111/j.1469-185X.2011.00208.x

Losos JB (2010) Adaptive radiation, ecological opportunity, and evolutionary determinism. Am Nat 175:623-639. doi: 10.1086/652433

Magnan P (1988) Interaction between Brook Charr, *Salvelinus fontinalis*, and nonsalmonid species: Ecological shift, morphological shift, and their impact on zooplankton communities. Can J Fish Aquat Sci 45:999-1009.

Magnan P, Stevens ED (1993) Pyloric caecal morphology of brook charr, *Salvelinus fontinalis*, in relation to diet. Environ Biol Fishes 36:205-210.

Magnan P, Proulx R, Plante M (2005) Integrating the effects of fish exploitation and interspecific competition into current life history theories: an example with lacustrine brook trout (*Salvelinus fontinalis*) populations. Can J Fish Aquat Sci 62:747-757. doi: 10.1139/F05-041

Magnusson A, Skaug H, Nielsen A, Berg C, Kristensen K, Maechler M, van Bentham K, Bolker B, Brooks M (2017) glmmTMB: generalized linear mixed models using template model builder – R package ver. 0.2.0.9. <https://github.com/glmmTMB>

Malmquist HJ (1992) Phenotype-specific feeding behaviour of two arctic charr *Salvelinus alpinus* morphs. Oecologia 92:354-361.

Martin RA, Pfennig DW (2010) Field and experimental evidence that competition and ecological opportunity promote resource polymorphism. Biol J Linn Soc 100:73-88. doi: 10.1111/j.1095-8312.2010.01380.x

Meyers SP (1994) Developments in world aquaculture, feed formulations, and role of carotenoids. Pure Appl Chem 66:1069-1076. doi: 10.1351/pac199466051069

Mittelbach GG (1981) Patterns of invertebrate size and abundance in aquatic habitats. Can J Fish Aquat Sci 38:896-904.

Mittelbach GG, Schemske DW, Cornell HV, Allen AP, Brown JM, Bush MB, Harrison SP, Hurlbert AH, Knowlton N, Lessios HA, McCain CM, McCune AR, McPeek MA, Near TJ, Price TD, Ricklefs RE, Roy K, Sax DF, Schluter D, Sobel JM, Turelli M (2007) Evolution and the latitudinal diversity gradient: speciation, extinction and biogeography. Ecol. Lett. 10:315-331.

Olsson J, Quevedo M, Colson C, Svanbäck R (2007) Gut length plasticity in perch: Into the bowels of resource polymorphisms. Biol J Linn Soc 90:517-523. doi: 10.1111/j.1095-8312.2007.00742.x

Parnell AC, Inger R, Bearhop S, Jackson AL (2010) Source partitioning using stable isotopes: Coping with too much variation. PLoS One 5:1-5. doi: 10.1371/journal.pone.0009672

Parnell A, Jackson A (2013) Stable isotope analysis in R (v.4.2) https://cran.r-project.org/web/packages/siar/index.html.

Pépino MP, Magnan P, Proulx R (2018) Field evidence for a rapid adaptive plastic response in morphology and growth of littoral and pelagic brook charr: a reciprocal transplant experiment. Funct Ecol 32:161-170.

Perga ME, Gerdeaux D (2005) "Are fish what they eat" all year round? Oecologia 144:598-606. doi: 10.1007/s00442-005-0069-5

Persson L (1987) Effects of habitat and season on competitive interactions between roach (*Rutilus rutilus*) and perch (*Perca fluviatilis*). Oecologia 73:170-177.

Pfennig DW, Wund MA, Snell-Rood EC, Cruickshank T, Schlichting CD, Moczek AP (2010) Phenotypic plasticity's impacts on diversification and speciation. Trends Ecol Evol 25:459-467. doi: 10.1016/j.tree.2010.05.006

Post DM (2002) Using stable isotopes to estimate trophic position: models, methods, and assumptions. Ecology 83:703-718.

Post DM, Layman CA, Arrington DA, Takimoto G, Quattrochi J, Montana CG (2007) Getting to the fat of the matter: models, methods and assumptions for dealing with lipids in stable isotope analyses. Oecologia 152:179-189. doi: 10.1007/s00442-006-0630-x

Pounder KC, Mitchell JL, Thomson JS, Pottinger TG, Sneddon LU (2018) Physiological and behavioural evaluation of common anaesthesia practices in the rainbow trout. App Anim Behav Sci 199: 94-102.

Proulx R, Magnan P (2004) Contribution of phenotypic plasticity and heredity to the trophic polymorphism of lacustrine brook charr (*Salvelinus fontinalis* M.). Evol Ecol Res 6:503-522.

Pysek P, Jarosik V, Hulme PE, Kuhn I, Wild J, Arianoutsou M, Bacher S, Chiron F, Didziulis V, Essl F, Genovesi P, Gherardi F, Hejda M, Kark S, Lambdon PW, Desprez-Loustau ML, Nentwig W, Pergl J, Poboljsaj K, Rabitsch W, Roques A, Roy DB, Shirley S, Solarz W, Vila M, Winter M (2010) Disentangling the role of environmental and human pressures on biological invasions across Europe. PNAS 107:12157-12162. doi: 10.1073/pnas.1002314107

Robinson BW, Wilson DS, Margosian AS, Lotito PT (1993) Ecological and morphological differentiation of pumpkinseed sunfish in lakes without bluegill sunfish. Evol Ecol 7:451-464. doi: 10.1007/BF01237641

Robinson BW, Wilson DS (1994) Character release and displacement in fishes: a neglected literature. Am Nat 144:596-627.

Robinson, BW, Schluter D (2000) Natural selection and the evolution of adaptive genetic variation in northern freshwater fishes. In: Mousseau T, Sinvero B, Endler JA (eds) Adaptive genetic variation in the wild. Oxford University Press, Oxford, pp. 65-94.

Robinson BW, Wilson DS, Margosian AS (2000) A pluralistic analysis of character release in pumpkinseed sunfish (*Lepomis gibbosus*). Ecology 81:2799-2812. doi: 10.2307/177342

Roesch C, Lundsgaard-Hansen B, Vonlanthen P, Taverna A, Seehausen O (2013) Experimental evidence for trait utility of gill raker number in adaptive radiation of a north temperate fish. J Evol Biol 26:1578-1587. doi: 10.1111/jeb.12166

Roughgarden J (1972) Evolution of niche width. Am Nat 106:683-718.

Rouleau S, Glémet H, Magnan P (2010) Effects of morphology on swimming performance in wild and laboratory crosses of brook trout ecotypes. Funct Ecol 24:310-321. doi: 10.1111/j.1365-2435.2009.01636.x

Saito A, Regier LW (1971) Pigmentation of brook trout (*Salvelinus fontinalis*) by feeding dried crustacean waste. J Fish Res Board Canada 28:509-512.

Schluter D (2000) The ecology of adaptive radiation. Oxford Univ. Press, Oxford.

Schneider RF, Meyer A (2017) How plasticity, genetic assimilation and cryptic genetic variation may contribute to adaptive radiations. Mol Ecol 26:330-350. doi: 10.1111/mec.13880

Skúlason S, Smith TB (1995) Resource polymorphisms in vertebrates. Trends Ecol Evol 10:366-370.

Skúlason S, Parsons KJ, Svanbäck R, Räsänen K, Ferguson MM, Adams CE, Amundsen PA, Bartels P, Bean CW, Boughman JW, Englund G, Guðbrandsson J, Hooker OE, Hudson AG, Kahilainen KK, Knudsen R, Kristjánsson BK, Leblanc CAL, Jónsson Z, Öhlund G, Smith C, Snorrason SS (2019) A way forward with eco evo devo: an extended theory of resource polymorphism with postglacial fishes as model systems. Biol Rev 94:1786-1808. doi: 10.1111/brv.12534

Smith TB, Skúlason S (1996) Evolutionary significance of resource polymorphisms in fishes, amphibians, and birds. Annu Rev Ecol Syst 27:111-133.

Strati IF, Sinanoglou VJ, Kora L, Miniadis-Meimaroglou S, Oreopoulou V (2012) Carotenoids from foods of plant, animal and marine origin: An efficient HPLC-DAD separation method. Foods 1:52-65. doi: 10.3390/foods1010052

Svanbäck R, Bolnick DI (2007) Intraspecific competition drives increased resource use diversity within a natural population. Proc Biol Sci 274:839-844. doi: 10.1098/rspb.2006.0198

Taylor EB, Bentzen P (1993) Evidence for multiple origins and sympatric divergence of trophic ecotypes of smelt (*Osmerus*) in northeastern North America. Evolution 47:813-832. doi: 10.1111/j.1558-5646.1993.tb01236.x

Tremblay S, Magnan P (1991) Interactions between two distantly related species, brook trout (*Salvelinus fontinalis*) and white sucker (*Catostomus commersoni*). Can J Fish Aquat Sci 48:857-867.

Trewby, ID, Wilson GJ, Delahay RJ, Walker N, Young R, Davison J, Cheeseman C, Robertson PA, Gorman ML, Mcdonald RA (2007) Experimental evidence of competitive release in sympatric carnivores. Biol Lett 4:170-172. doi: 10.1098/rsbl.2007.0516

Vander Zanden J, Rasmussen JB (1999) Primary consumer δ13C and δ15N and the trophic position of aquatic consumers. Ecol Monogr 80:1395-1404.

Wellborn GA, Langerhans RB (2015) Ecological opportunity and the adaptive diversification of lineages. Ecol Evol 5:176-195. doi: 10.1002/ece3.134. 7

Wetzel RG, Likens GE (2000) Limnological analyses, 3rd ed. Springer-Verlag, New York.

Yoder, JB, Clancey E, Des Roches S, Eastman JM, Gentry L, Godsoe W, Hagey TJ, Jochimsen D, Oswald BP, Robertson J, Sarver BAJ, Schenk JJ, Spear SF, Harmon LJ (2010) Ecological opportunity and the origin of adaptive radiations. J Evol Biol 23:1581-1596. doi: 10.1111/j.1420-9101.2010.02029.x

Zaccarelli N, Bolnick DI, Mancinelli G (2013) RInSp: an r package for the analysis of individual specialization in resource use. Methods Ecol Evol 4:1018-1023. doi: 10.1111/2041-210X.12079

Tables

Table 1. Lake characteristics and sample sizes used in statistical analysis of the 27 study lakes. For all indicators, the number of brook charr samples is presented for both the littoral and pelagic zones (L/P). Sampling date indicates the first day of sampling; sampling lasted four (2013, 2014) or five (2012) days.

Lake	Sampling dates	Fish community*	Area (HA)	Mean depth (m)	Littoral zone proportion (%)	Mean competitor biomass (g/trap)	Mean brook charr biomass g/trap (L/P)	Stomach content (L/P)	Carbon signature (L/P)	Astaxanthin (L/P)	SalmoFan (L/P)	Caecum (L/P)	Gill Raker (L/P)
Baie des Onze Îles (A)	19-Jun + 26-Jul-2012		135	7.20	7	0	181/517	23/47	18/19	-	-	-	-
Baie Verte (B)	26-Jun + 1-Aug-2012		-	-	-	0	148/1026	14/53	15/20	-	-	-	-
Baie Cobb (C)	03 + 20-Jul-2012		62	5.88	8	0	286/896	33/55	19/19	-	-	-	-
Maréchal (D)	10 + 17-Jul-2012		103	5.18	10	0	508/3797	33/46	20/18	-	-	-	-
Vertnez (E)	20-May-2013	SEAT	-	-	-	16	2683/401	39/34	19/20	-	-	-	-
Bucheron (G)	03-Jun-2013		10	3.90	18	0	2275/345	31/51	20/20	-	-	-	5/5
Cutaway (H)	10-Jun-2013	CHEO, SEAT, CACO	40	3.38	15	312	1885/850	21/25	-	-	-	-	5/4
Camp (I)	17-Jun-2013	CHEO, SEAT, CACO	11	5.82	13	505	1453/207	15/7	20/10	-	-	-	5/5
Visons (J)	24-Jun-2013	CHEO, SEAT	74	7.54	6	12	939/1533	37/30	-	-	-	-	-
Jimmy (K)	02-Jul-2013	CHEO, SEAT, CACO, LUCO	-	-	-	169	910/1030	13/12	-	10/10	54/37	-	-

Table 1 (Continued)

Lake	Sampling dates	Fish community*	Area (HA)	Mean depth (m)	Littoral zone proportion (%)	Mean competitor biomass (g/trap)	Mean brook charr biomass g/trap (L/P)	Stomach content (L/P)	Carbon signature	Lake	Sampling dates	Fish community*	Area (HA)
Brise (L)	08-Jul-2013	CHEO, SEAT, CACO	39	3.59	23	304	64/334	11/29	16/20	10/9	16/38	-	5/5
Lézard (M)	15-Jul-2013	CHEO, SEAT, CACO	110	7.83	5	469	0/167	0/8	-	-	0/10	-	-
Chute Noire (N)	22-Jul-2013	CHEO	17	3.80	18	0	1120/591	28/24	22/29	10/9	60/59	-	5/5
Corneille (O)	29-Jul-2013	CHEO, SEAT, CACO	23	4.78	17	347	316/1241	15/46	20/20	10/9	26/60	-	4/5
Bourassa (P)	05-Aug-2013	CHEO	56	7.26	8	0	565/2587	20/37	15/20	10/10	40/45	-	3/5
Coteau (Q)	20-May-2014	CHEO	31	4.72	16	0	3137/499	32/46	21/19	-	-	15/14	15/14
Oudiette (R)	27-May-2014	CHEO, SEAT, CACO	44	8.59	7	211	1227/336	36/23	20/20	-	55/36	14/15	15/15
Gauthier (S)	03-Jun-2014	CHEO, SEAT, MAMA	37	8.62	9	22	1327/781	27/27	18/20	-	53/55	15/13	15/13
Shawinigan (T)	10-Jun-2014	CHEO, SEAT, LUCO, MAMA	60	11.70	4	26	1221/739	27/23	16/19	-	48/53	12/15	12/15
Adam (U)	17-Jun-2014		14	5.24	13	0	1651/1229	47/47	20/18	-	59/57	13/12	15/15

Table 1 (Continued)

Lake	Sampling dates	Fish community*	Area (HA)	Mean depth (m)	Littoral zone proportion (%)	Mean competitor biomass (g/trap)	Mean brook charr biomass g/trap (L/P)	Stomach content (L/P)	Carbon signature(L/P)	Astaxanthin (L/P)	SalmoFan (L/P)	Caecum (L/P)	Gill Raker (L/P)
Dorval (V)	24-Jun-2014	CHEO, CACO, COPL, CUIN, RHCA	-	-	-	285	53/182	0/2	-	-	0/5	1/5	1/5
Marshall (W)	01-Jul-2014	SEAT	41	6.39	10	48	855/1330	39/49	20/20	-	58/59	15/15	15/15
Marmotte (X)	08-Jul-2014	SEAT	14	2.97	27	38	921/2407	23/51	19/20	-	56/55	15/15	15/15
Longue Vue (Y)	15-Jul-2014	SEAT	28	5.44	16	18	2897/1890	43/56	20/20	-	56/58	14/15	14/15
Plouf (Z)	22-Jul-2014	CHEO, SEAT, CACO, PIPR	60	8.82	5	378	633/1230	22/27	18/20	-	49/58	15/15	15/14
Thibert (AA)	29-Jul-2014	SEAT	43	5.79	8	28	335/808	20/28	20/19	-	35/59	15/14	15/13
Simpson (BB)	05-Aug-2014		30	4.24	26	0	1280/1387	39/54	17/17	-	53/61	13/15	13/15
Total:								**688/937**	**413/427**	**50/47**	**718/815**	**157/163**	**192/198**

* Fish species codes: SEAT, creek chub (*Semotilus atromaculatus*); CHEO, northern redbelly dace (*Chrosomus eos*); CACO, white sucker (*Catostomus commersonii*); LUCO, common shiner (*Luxilus cornutus*); MAMA, Allegheny pearl dace (*Margariscus margarita*); COPL, lake chub (*Couesius plumbeus*); CUIN, brook stickleback (*Culaea inconstans*); RHCA, longnose dace (*Rhinichthys cataractae*); and PIPR, fathead minnow (*Pimephales promelas*). Brook charr were present in all lakes.

Table 2. Comparison of 11 candidate models differing in their fixed effect components for five indicators of resource use in littoral and pelagic zones by brook charr in 27 Canadian Shield lakes. The number of model parameters (K) and difference in AICc relative to the best-fitting model (ΔAICc) are presented for all indicators except for carbon stable isotope, for which we show the percentage of time a model was selected as the best after 1000 iterations. Each model included standardized log-transformed fish length (L) and standardized day of the year (D) as fixed effects. The lake and year of sampling were included as random effects in all models except for astaxanthin concentration and caecum length models, where only the lake was included (all data are from 2014). All models predicting muscle colour (Roche SalmoFan© lineal card) also include the observer as random effect (two different observers). For each indicator, the best selected model is shown in bold.

Model	Terms in the model[a]	K	Stomach content (ΔAICc)	Carbon stable isotope (% of selection)	Carotenoids		Ceacum length (ΔAICc)[b]	Gill raker length (ΔAICc)
					Astaxanthin concentration (ΔAICc)[b]	Muscle colour (ΔAICc)[c]		
1	Null model (L+D)	6	48.7	0	6.6	5.7	12.5	4.7
2	Z	7	12.6	**75.1**	2.4	0.9	9.5	6.1
3	S	7	49.1	0	8.9	5.9	14.6	6.0
4	C	8	44.8	0	8.1	4.8	2.9	**0.0**
5	Z + S	8	12.9	1.3	4.7	**1.1**	11.6	7.4
6	Z × S	9	14.4	5.4	5.0	1.2	10.0	5.6
7	C + S	9	45.0	0	10.4	5.0	5.0	1.4
8	C × S	11	47.9	0	12.8	6.3	6.6	3.6
9	C + Z	9	7.7	4.9	3.8	0.0	**0.0**	1.4
10	C × Z	11	8.1	0.5	5.6	4.0	4.0	1.3
11	C × D + Z	11	**0.0**	12.8	**0.0**	3.8	1.5	3.8

a) Model terms are Z: zone of capture (littoral or pelagic); S: sex (male or female); C: fish community (brook charr only, brook charr in sympatry with creek chub, or with creek chub and white sucker); D: day of the year.

b) Because we did not include the year as a random effect for all astaxanthin and caecum length models (all data from 2014), the model parameters (K) are lower by one unit. The fish community variable (C) has only two levels in all astaxanthin concentration models, which also lowers K by one unit for models 4, 7, and 9, and two units for models 8, 10, and 11.

c) Because observer was included as a random effect in all SalmoFan models, the number of model parameters (K) is one unit higher.

Table 3. Main statistics of the best competing models for stomach content, carbon stable isotope, astaxanthin concentration, muscle colour, ceacum length, and gill raker length of brook charr. C: fish community; D: day of the year (standardized); L: total fish length (log-transformed and standardized); Z: zone of capture. The carbon stable isotope model is based on the mean individual pelagic contribution from the mixing models (see text).

Indicator	Estimate	Std. Error	Z value	t value	Variance	SD
Stomach content (N = 1625; 27 lakes)						
Intercept	-0.53489	0.13116	-4.078	-	-	-
C: BC+CC	0.45197	0.19790	2.284	-	-	-
C: BC+CC+WS	0.69800	0.20463	3.411	-	-	-
D	-0.08629	0.10200	-0.846	-	-	-
D × C: BC+CC	0.55437	0.18906	2.932	-	-	-
D × C: BC+CC+WS	0.58043	0.20024	2.899	-	-	-
L	-0.16461	0.03576	-4.603	-	-	-
Z	0.20836	0.03411	6.108	-	-	-
Lake (Random effect)	-	-	-	-	1.423e-01	3.772e-01
Year (Random effect)	-	-	-	-	9.053e-09	9.514e-05
Overdispersion	0.41					
Carbon stable isotope (N = 840; 22 lakes)						
Intercept	-1.16233	0.0913	-12.73	-	-	-
D	0.03418	0.07798	0.438	-	-	-
L	-0.08382	0.02446	-3.427	-	-	-
Z	0.18098	0.0913	7.863	-	-	-
Lake (Random effect)	-	-	-	-	0.1705	0.4129
Overdispersion	10.8					
Astaxanthin concentration (N = 97; 5 lakes)						
Intercept	0.30255	0.28981	-	1.044	-	-
C: BC+CC+WS	0.10937	0.32939	-	0.332	-	-
D	-0.83128	0.3158	-	-2.632	-	-

Table 3 (Continued)

Indicator	Estimate	Std. Error	Z value	t value	Variance	SD
L	0.04132	0.09448	-	0.437	-	-
Z	0.22297	0.08647	-	2.579	-	-
Lake (Random effect)	-	-	-	-	0.02098	0.1448
Residual	-	-	-	-	0.70487	0.8396
Muscle color (N = 1533; 17 lakes)						
Intercept	0.10566	0.20920	-	0.505	-	-
D	-0.03920	0.17627	-	-0.222	-	-
L	0.27111	0.01988	-	13.638	-	-
Z	0.05220	0.01945	-	2.683	-	-
Lake (Random effect)	-	-	-	-	0.57-393	0.7576
Observer (Random effect)	-	-	-	-	0.03746	0.1936
Residual	-	-	-	-	0.52498	0.7246
Ceacum length (N = 320; 12 lakes)						
Intercept	-0.30650	0.11705	-	-2.618	-	-
C: BC+CC	0.27286	0.14339	-	1.903	-	-
C: BC+CC+WS	0.81342	0.17384	-	4.679	-	-
D	-0.05426	0.06051	-	-0.897	-	-
L	0.63170	0.03868	-	16.332	-	-
Z	0.08535	0.03806	-	2.242	-	-
Lake (Random effect)	-	-	-	-	0.02388	0.1545
Residual	-	-	-	-	0.46019	0.6784

Table 3 (Continued)

Indicator	Estimate	Std. Error	Z value	t value	Variance	SD
Gill raker length (N = 390; 19 lakes)						
Intercept	-0.23421	0.09856	-	-2.376	-	-
C: BC+CC	0.31105	0.13282	-	2.342	-	-
C: BC+CC+WS	0.40380	0.13860	-	2.342	-	-
D	-0.04227	0.05526	-	-0.765	-	-
L	0.72875	0.03278	-	22.228	-	-
Lake (Random effect)	-	-	-	-	0.03419	0.1849
Residual	-	-	-	-	0.38230	0.6183

Table 4. Estimates of the model comparing HPLC astaxanthin concentration and muscle colour estimated using the SalmoFan© card. The model is based on linear regression of 96 observations. The multiple R^2 is 0.362. HPLC values were log transformed.

Fixed effect	Estimate	Std. Error	t value	p-value
Intercept	-0.71380	0.61957	-1.152	0.2522
Log(SalmoFan)	0.11591	0.02356	4.921	< 0.001
Observer (V)	-0.50185	0.13432	-3.736	< 0.001

Figures captions

Figure 1. Roche SalmoFan© lineal card used to evaluate brook charr muscle colour. Muscle samples are from Lake Maréchal, La Mauricie National Park (Québec, Canada).

Figure 2. Mean percent dry weight of each prey category in the stomach content of 1625 brook charr. a) Lakes with brook charr only (n = 10), b) lakes with brook charr and creek chub (n = 8), and c) lakes with brook charr, creek chub, and white sucker (n = 9).

Figure 3. Marginal model effects (mean ± 95% CI) for fish community and/or the zone of capture (littoral, pelagic) on the a) proportion of pelagic prey in stomach contents, b) relative contribution of pelagic prey to $\delta^{13}C$ in liver, c) astaxanthin concentration in muscle (mg/kg), d) muscle colour (Roche SalmoFan© lineal card), e) posterior caecum length (mm), and f) gill raker length (mm) of brook charr. The best model is represented by circles. For comparative purposes, we also represented the effects of fish community and zone of capture with squares when not present in the best model. Zone of capture: littoral (L) symbols are red and pelagic (P) are blue. Fish community: brook charr lakes (BC); brook charr and creek chub lakes (BC+CC); and brook charr, creek chub and white sucker lakes (BC+CC+WS). The grey dashed line in (a) represents equal proportions of benthic and pelagic prey.

Figure 4. Frequency distributions of fish according to the percentage of pelagic prey (stomach content; dry weight) for brook charr (BC), brook charr and creek chub (BC+CC), and brook charr, creek chub, and white sucker (BC+CC+WS) lakes. The number of lakes appears in parentheses. Ecotype (based on the zone of capture): littoral in red and pelagic in blue. The number of stomach contents analyzed was 760 for BC lakes, 553 for BC+CC lakes, and 310 for CB+CC+WS lakes.

Figure 5. Estimates (black lines) ± SD (dark grey area) and 2 × SD (light grey area) of the best isotopic model for 1000 iterations. The mean estimates ± mean SD of the

1000 iterations was -1.187 ± 0.152 for the intercept, 0.178 ± 0.029 for the zone (effect coding), -0.084 ± 0.031 for fish length, and -0.051 ± 0.073 for day of the year.

Figure 6. Probability distributions of the pelagic contribution to liver $\delta^{13}C$ based on stable isotope analysis in an R mixing model that included all BC lakes. The distributions are based on 1000 estimations for each individual. The blue and red squares are the mean estimations of pelagic and littoral individuals, respectively, based on their zone of capture. The blue and red vertical lines represent the 95% confidence interval. See Table 1 for lake abbreviations.

Figure 7. Probability distributions of the pelagic contribution to liver $\delta^{13}C$ based on stable isotope analysis in an R mixing model that included all BC+CC lakes. The distributions are based on 1000 estimations for each individual. The blue and red squares are the mean estimations of pelagic and littoral individuals, respectively, based on their zone of capture. The blue and red vertical lines represent the 95% confidence interval. See Table 1 for lake abbreviations.

Figure 8. Probability distributions of the pelagic contribution to the liver $\delta^{13}C$ based on stable isotope analysis in an R mixing model that included all BC+CC+WS lakes. The distributions are based on 1000 estimations for each individual. The blue and red squares are the mean estimations of pelagic and littoral individuals, respectively, based on their zone of capture. The blue and red vertical lines represent the 95% confidence interval. See Table 1 for lake abbreviations.

Figure 9. Linear regression between astaxanthin concentration (mg/kg) from HPLC and muscle colour from Roche SalmoFan© lineal card for 96 brook charr muscle samples. The open circles and dashed line represent samples measured by observer M while the grey circles and solid line represent samples measured by observer V. The Y axis is expressed on a log scale.

Figure 10. Individual (WIC) vs. population (TNW) niche widths of the 25 brook charr lake populations considered in this analysis. Fish community: brook charr lakes (BC; blue); brook charr and creek chub lakes (BC+CC; orange); and brook charr, creek chub, and white sucker lakes (BC+CC+WS; red). Because TNW = WIC+BIC (BIC: between-individual component of TNW), all populations must fall on or below the solid black line, which represents the 1:1 ratio (i.e., when WIC = TNW). The thin dashed grey lines represent increasing isoclines of individual specialization (smaller WIC/TNW; Bolnick et al., 2010; see text). Both WIC and TNW were log-transformed for better visualization.

Figure 11. Distributions of (a) individual (WIC) and (b) population (TNW) niche widths of the 25 brook charr lake populations considered in this analysis. Fish community: brook charr lakes (BC); brook charr and creek chub lakes (BC+CC); and brook charr, creek chub, and white sucker lakes (BC+CC+WS). Boxplots with mean values, 25^{th} and 75^{th} percentiles, and whiskers representing the 10^{th} and 90^{th} percentiles are shown; outliers are represented by dots. Both WIC and TNW are on a log scale.

Figures

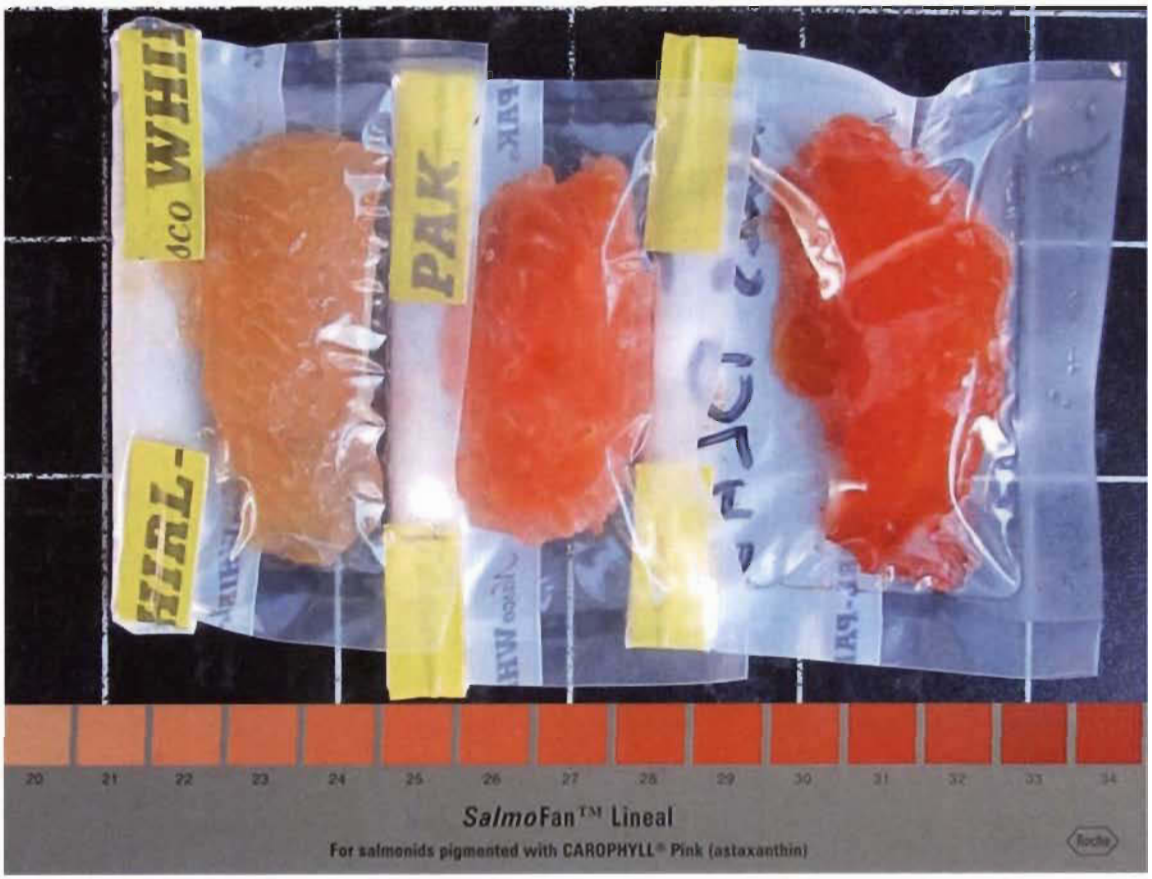

Figure 1.

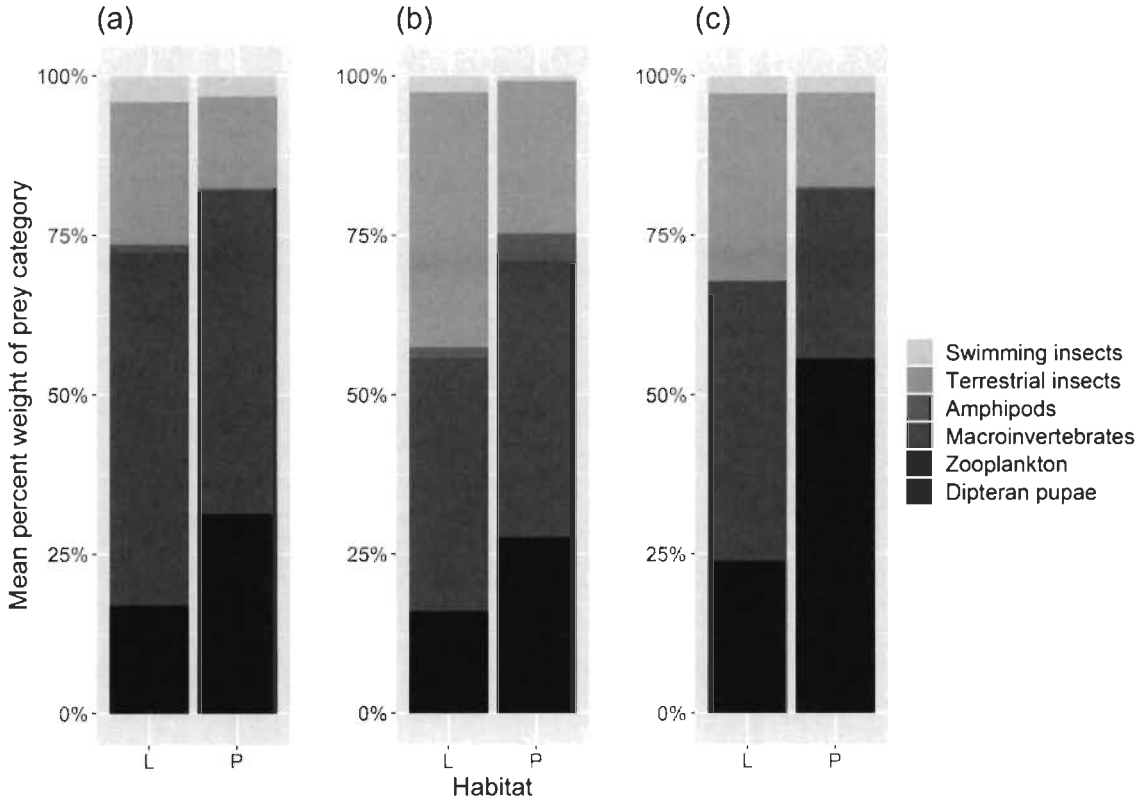
(a)
(b)
(c)
100%
75%
50%
25%
0%
Mean percent weight of prey category
L
P
Habitat
Swimming insects
Terrestrial insects
Amphipods
Macroinvertebrates
Zooplankton
Dipteran pupae

Figure 2.

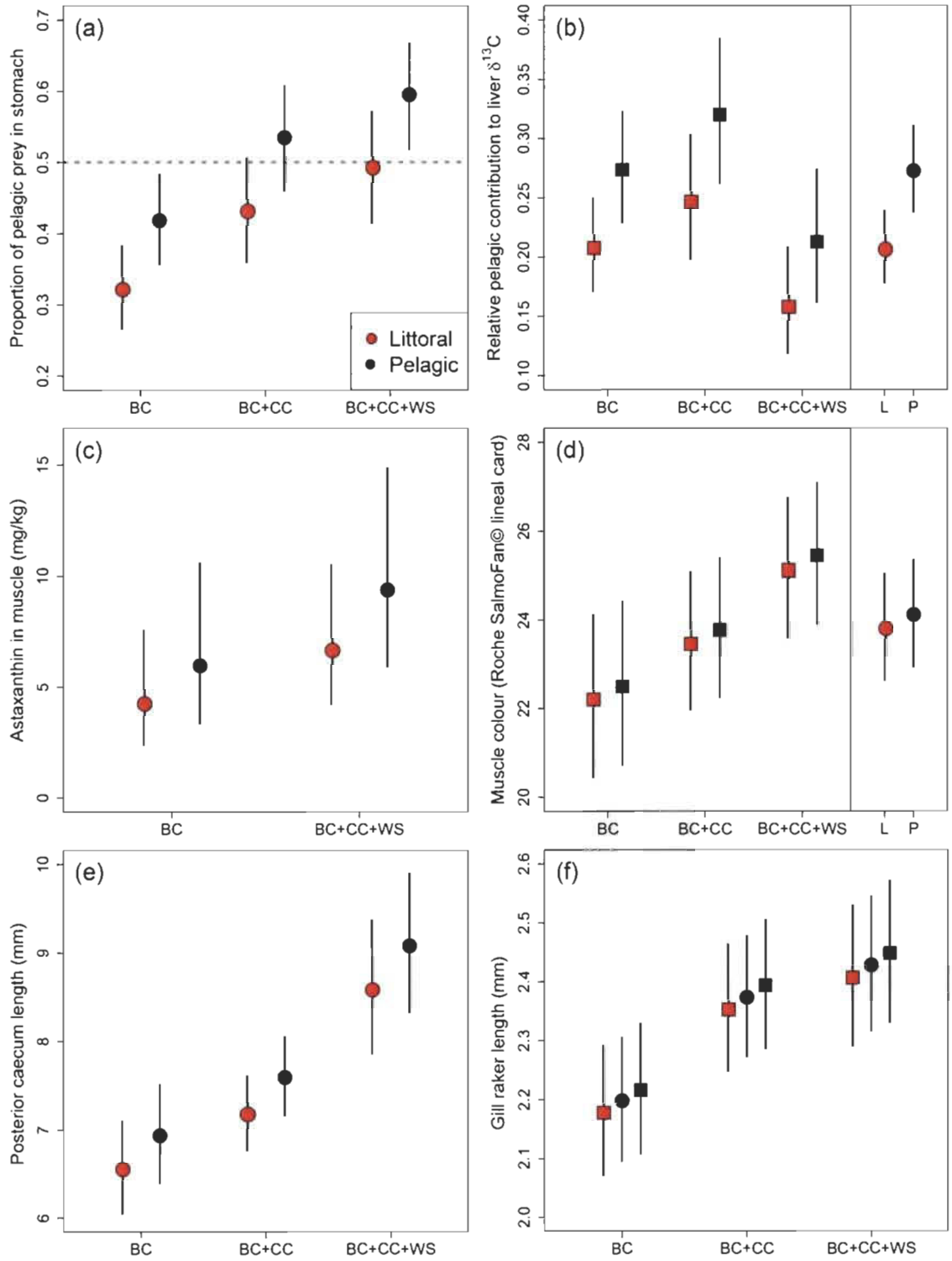
(a)
Proportion of pelagic prey in stomach
0.2
0.3
0.4
0.5
0.6
0.7
Littoral
Pelagic
BC
BC+CC
BC+CC+WS
(b)
Relative pelagic contribution to liver δ13C
0.10
0.15
0.20
0.25
0.30
0.35
0.40
BC
BC+CC
BC+CC+WS
L
P
(c)
Astaxanthin in muscle (mg/kg)
0
5
10
15
BC
BC+CC+WS
(d)
Muscle colour (Roche SalmoFan© lineal card)
20
22
24
26
28
BC
BC+CC
BC+CC+WS
L
P
(e)
Posterior caecum length (mm)
6
7
8
9
10
BC
BC+CC
BC+CC+WS
(f)
Gill raker length (mm)
2.0
2.1
2.2
2.3
2.4
2.5
2.6
BC
BC+CC
BC+CC+WS

Figure 3.

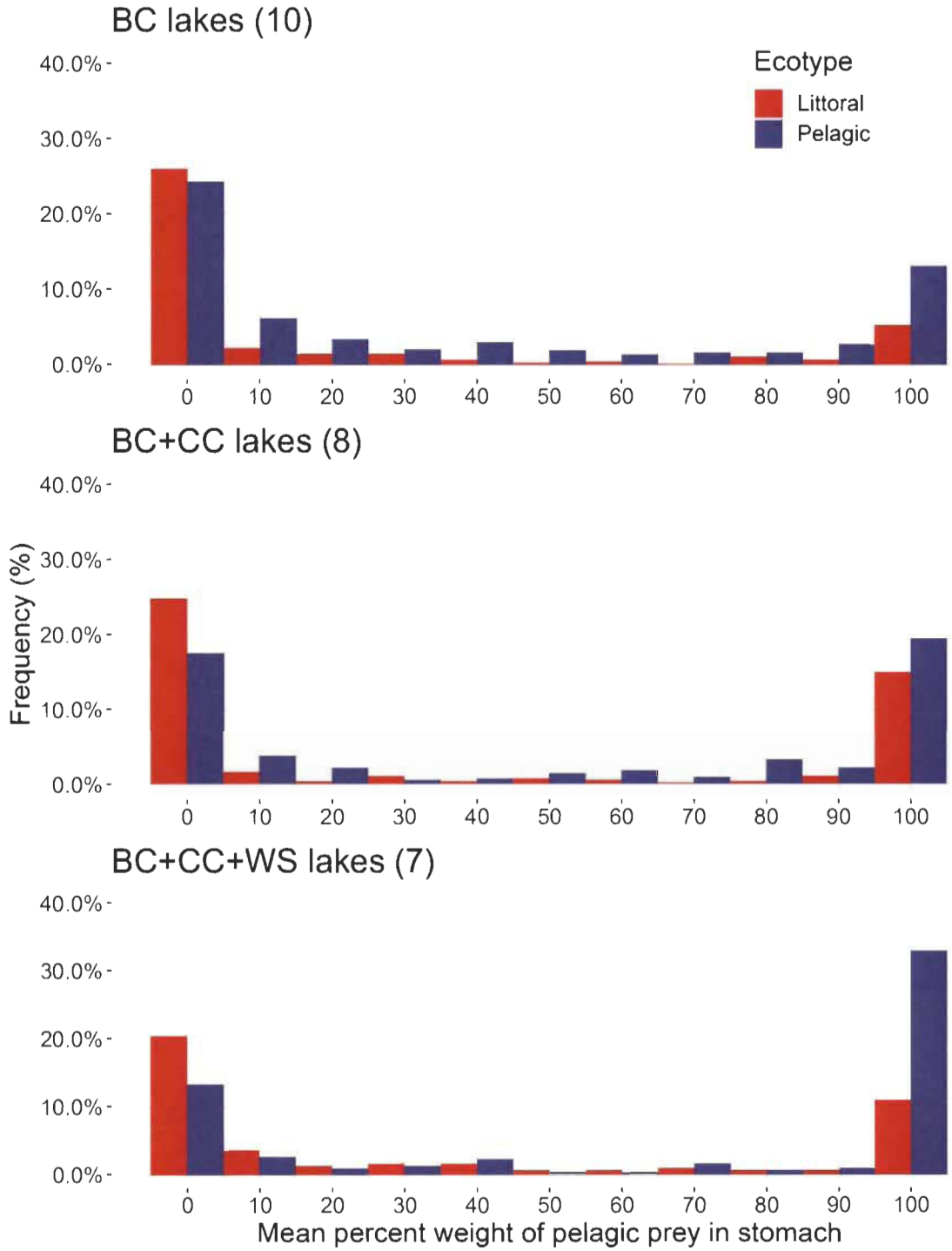
BC lakes (10)
BC+CC lakes (8)
BC+CC+WS lakes (7)
Ecotype
Littoral
Pelagic
Frequency (%)
40.0%
30.0%
20.0%
10.0%
0.0%
0
10
20
30
40
50
60
70
80
90
100
Mean percent weight of pelagic prey in stomach

Figure 4.

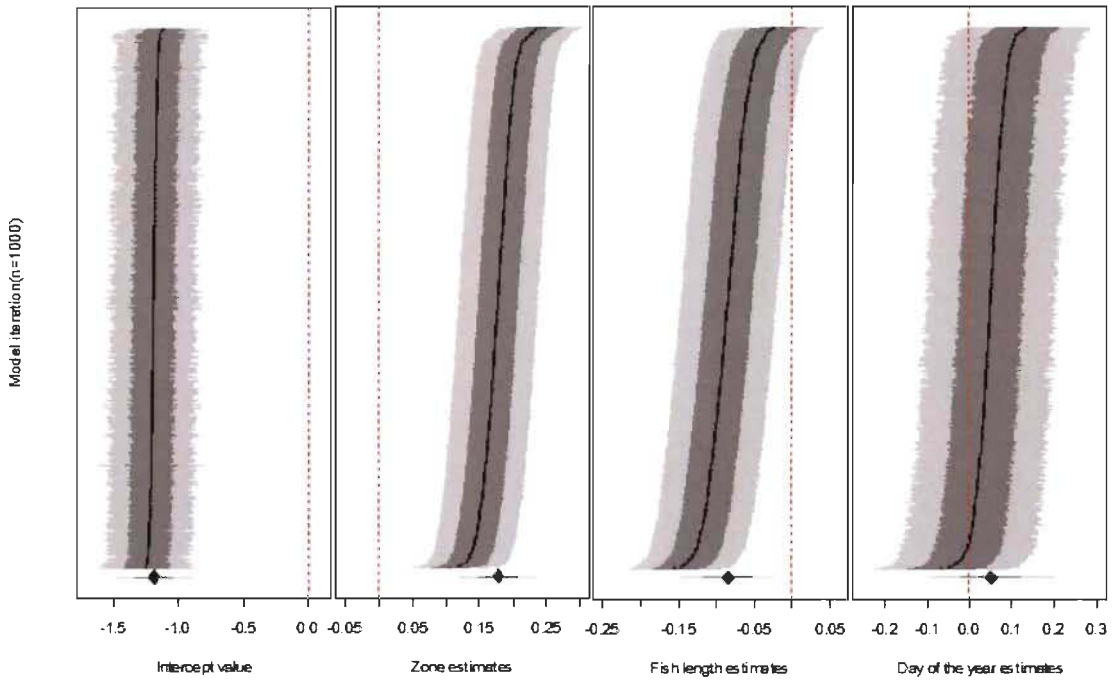
Model iteration(n=1000)
-1.5
-1.0
-0.5
0.0
Intercept value
-0.05
0.05
0.15
0.25
Zone estimates
-0.25
-0.15
-0.05
0.05
Fish length estimates
-0.2
-0.1
0.0
0.1
0.2
0.3
Day of the year estimates

Figure 5.

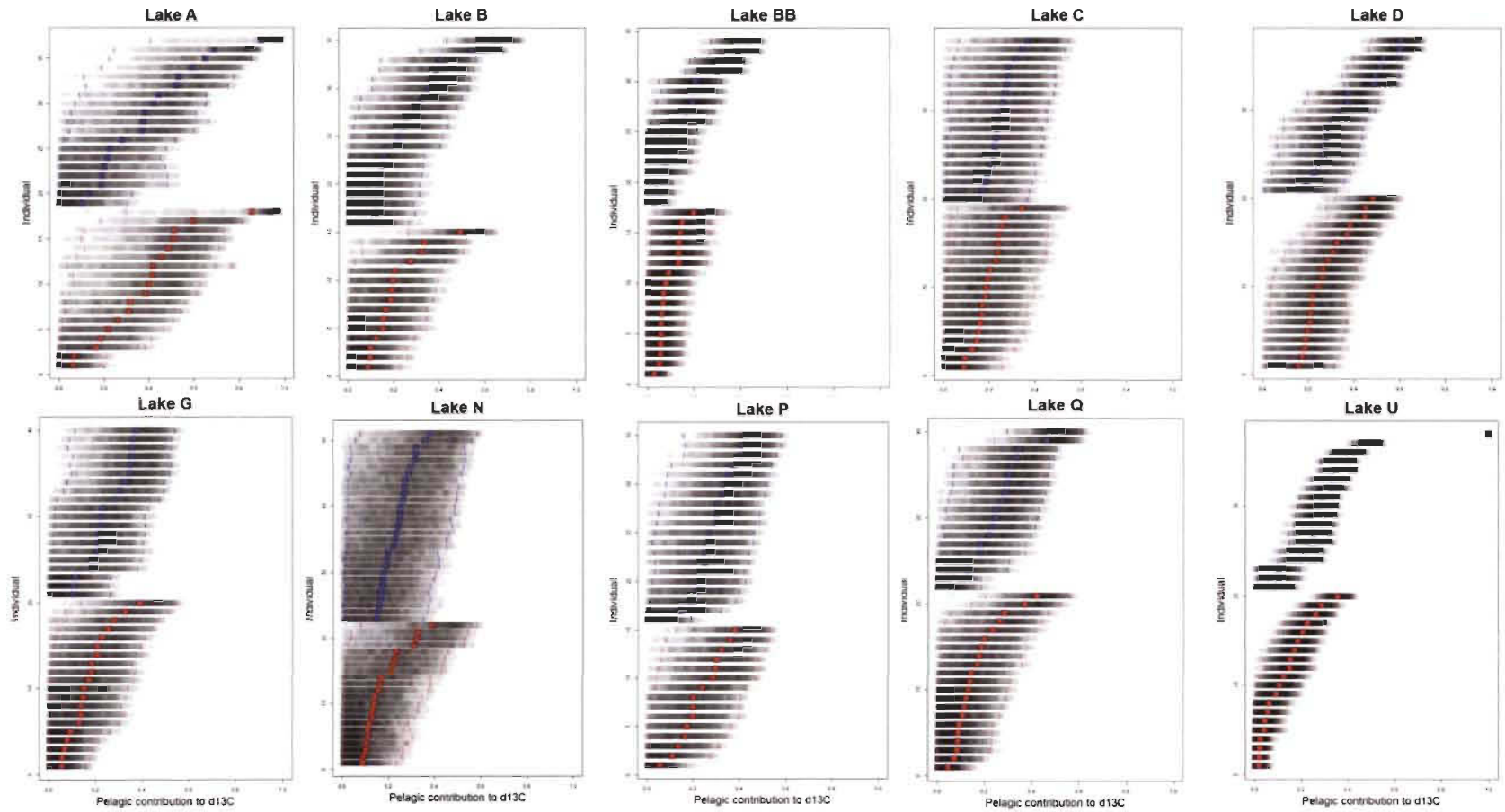
Lake A
Lake B
Lake BB
Lake C
Lake D
Lake G
Lake N
Lake P
Lake Q
Lake U
Individual
Pelagic contribution to d13C

Figure 6.

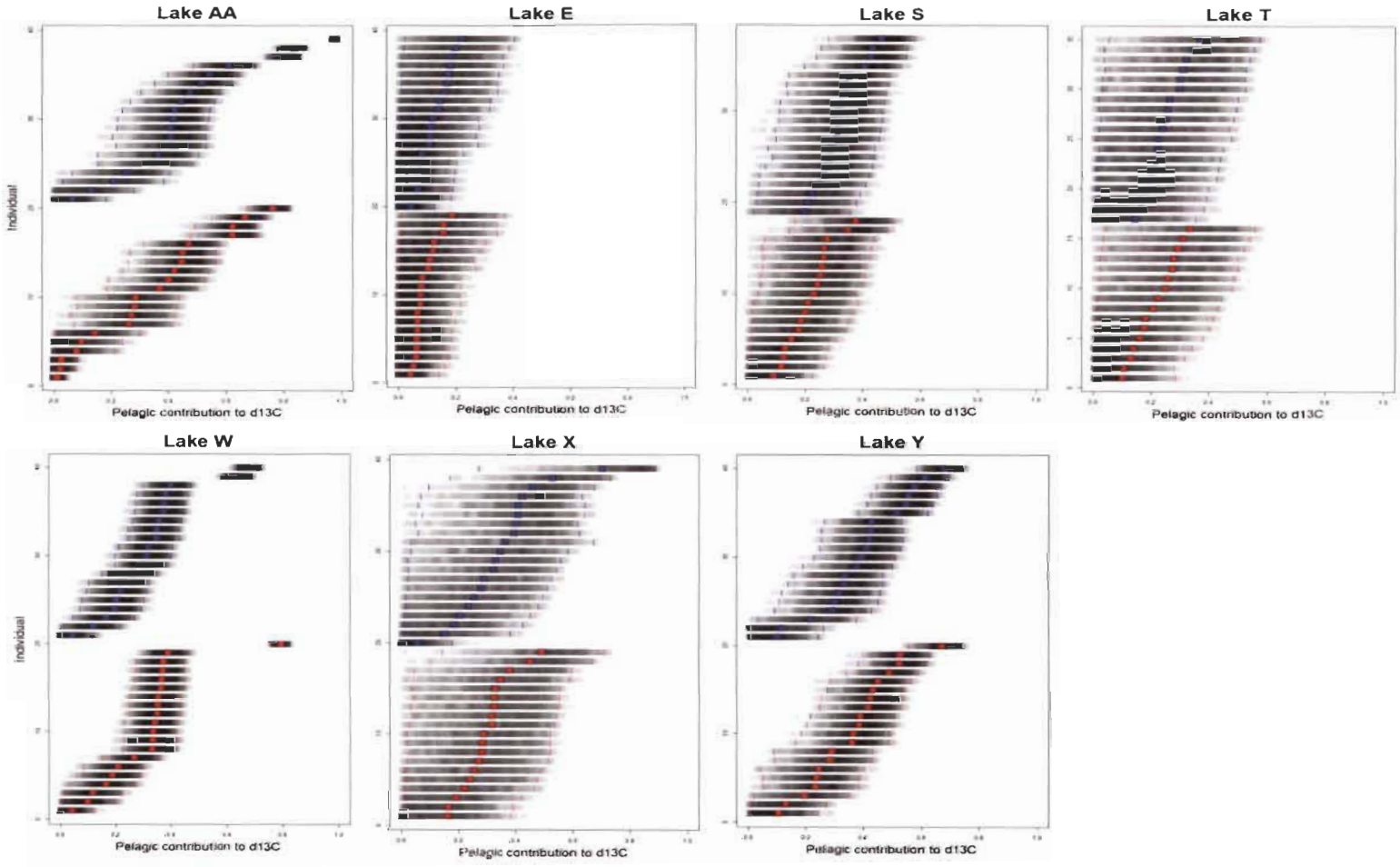
Lake AA
Lake E
Lake S
Lake T
Lake W
Lake X
Lake Y
Individual
Pelagic contribution to d13C

Figure 7.

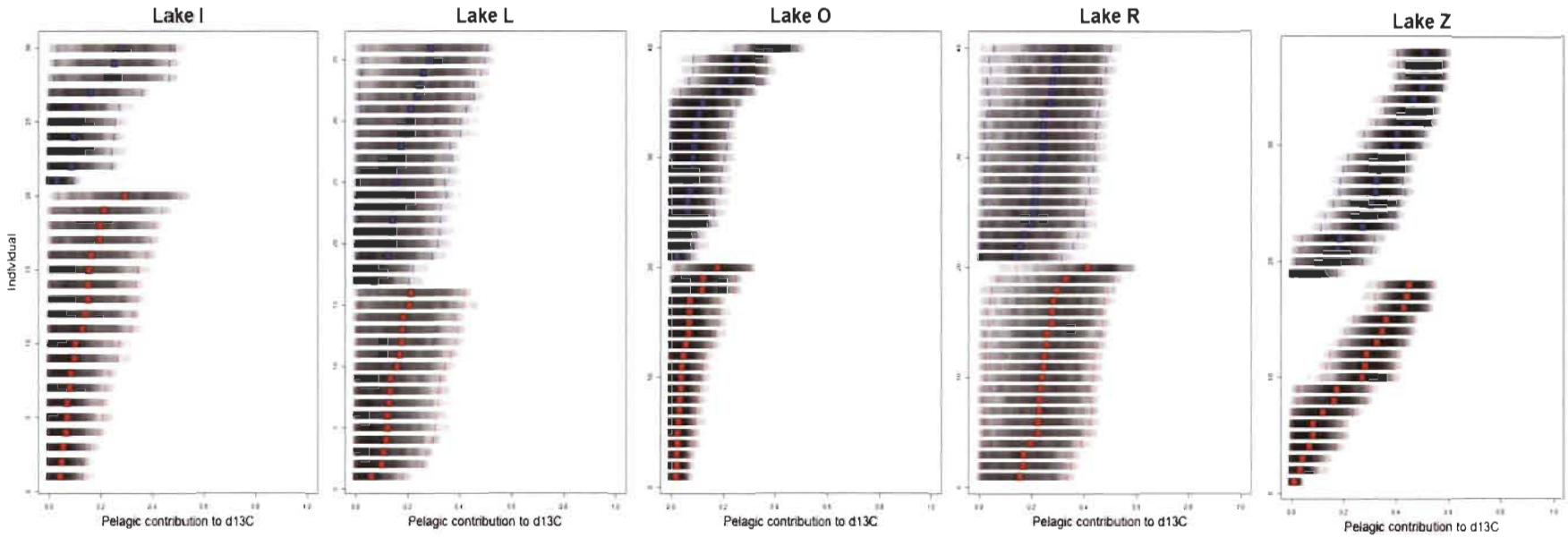
Lake I
Lake L
Lake O
Lake R
Lake Z
Individual
Pelagic contribution to d13C
Pelagic contribution to d13C
Pelagic contribution to d13C
Pelagic contribution to d13C
Pelagic contribution to d13C

Figure 8.

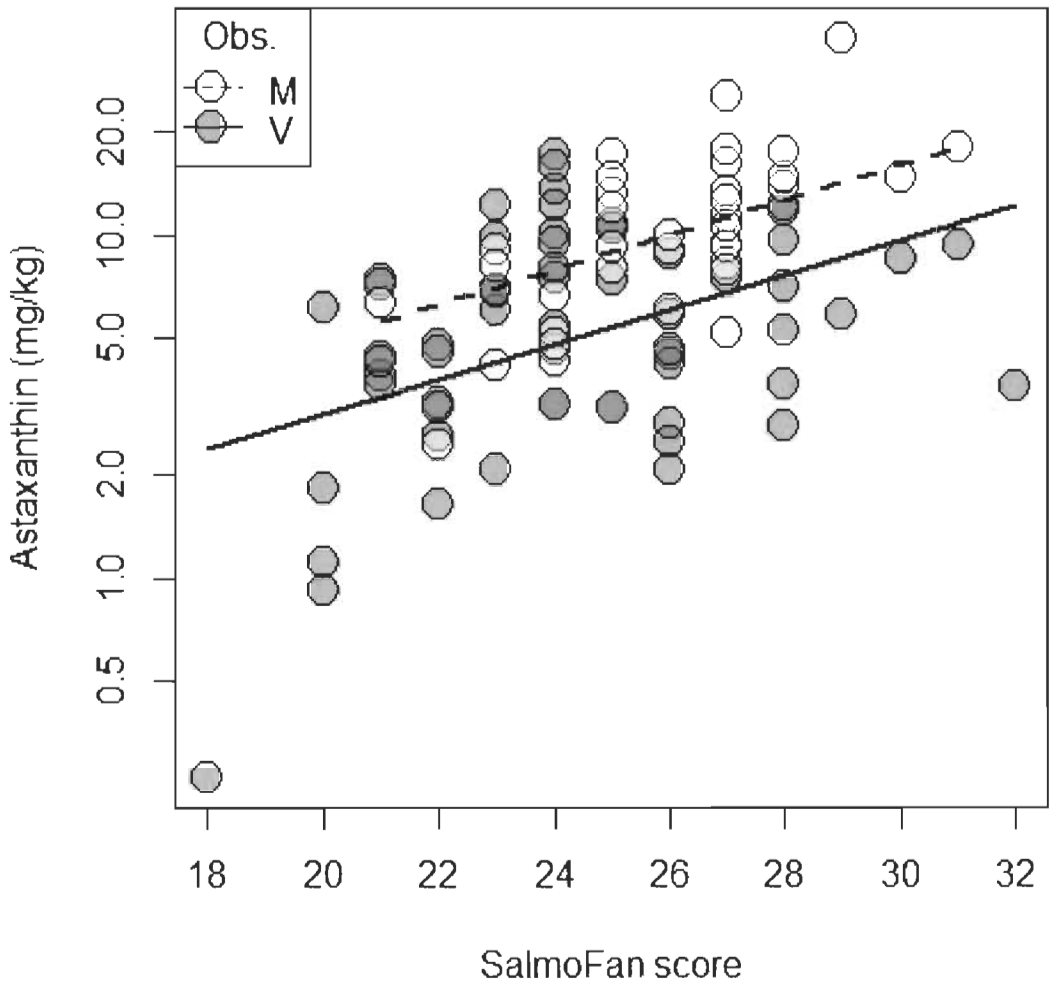
Obs.
M
V
Astaxanthin (mg/kg)
0.5
1.0
2.0
5.0
10.0
20.0
18
20
22
24
26
28
30
32
SalmoFan score

Figure 9.

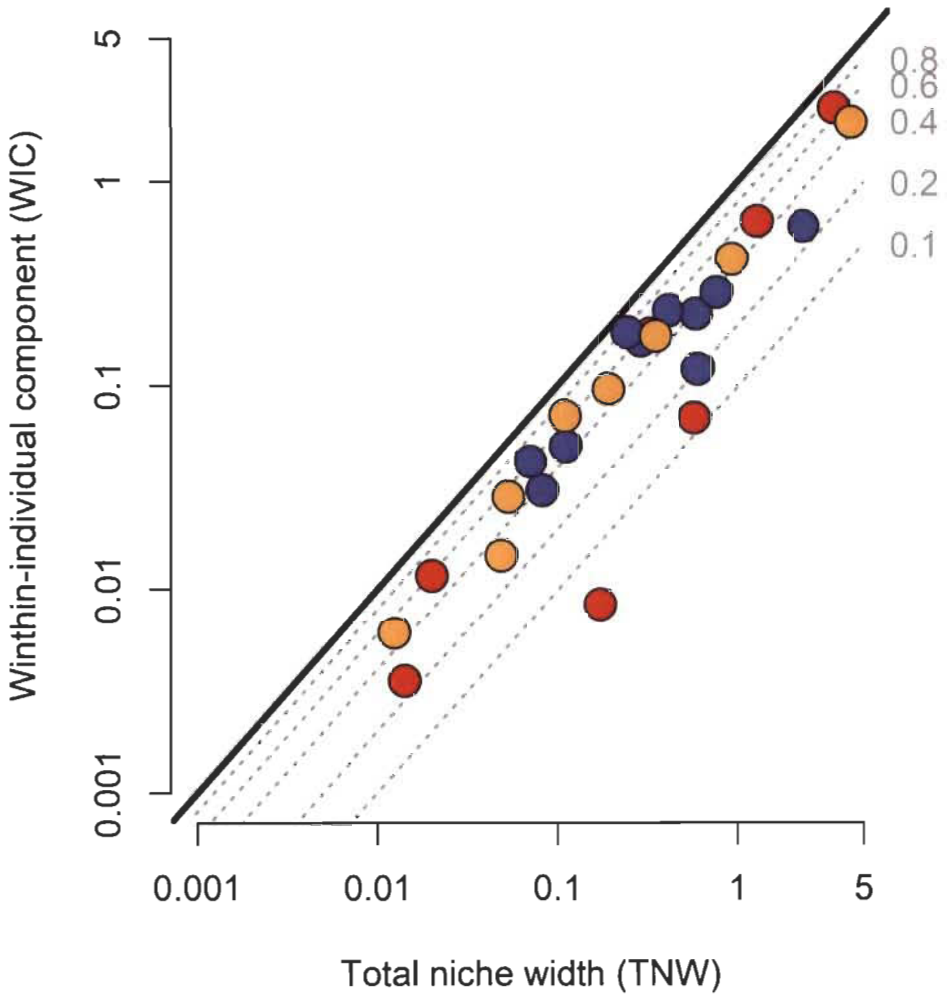
Winthin-individual component (WIC)
0.001
0.01
0.1
1
5
Total niche width (TNW)
0.001
0.01
0.1
1
5
0.8
0.6
0.4
0.2
0.1

Figure 10.

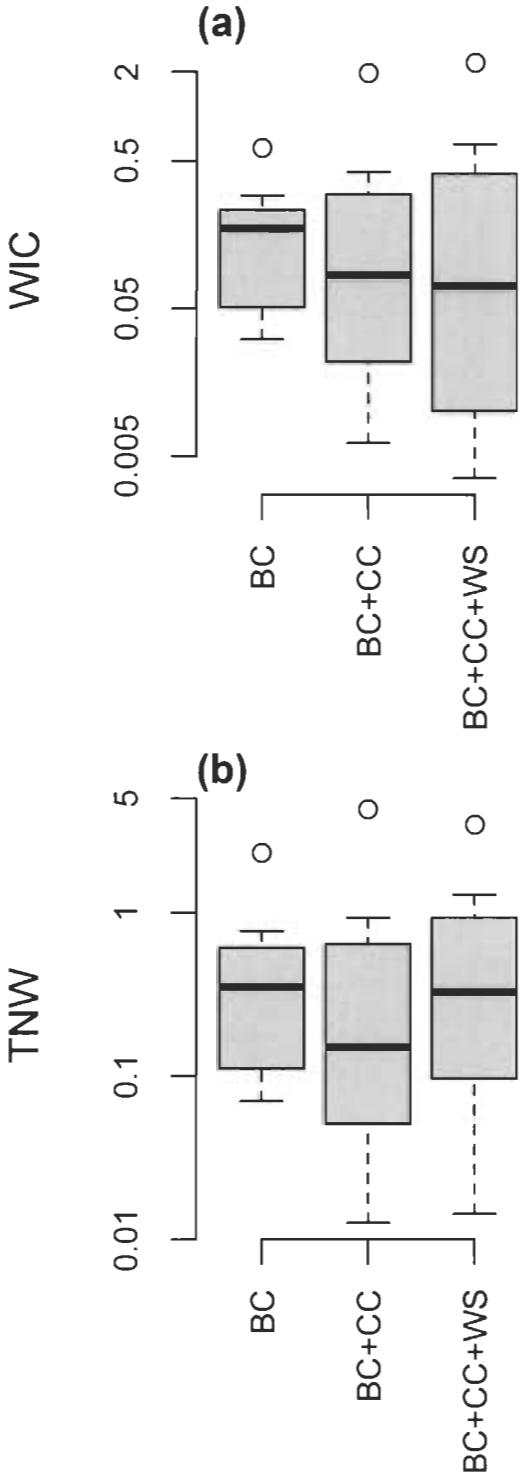

Figure 11.

CHAPITRE IV

PARALLEL EVOLUTION OF MORPHOLOGICAL TRAITS AND BODY SHAPE IN LITTORAL AND PELAGIC BROOK CHARR, *SALVELINUS FONTINALIS*, ALONG A GRADIENT OF INTERSPECIFIC COMPETITION

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Author contributions

PM originally formulated the idea. VR, MP conducted fieldwork. VR wrote the first draft of the manuscript. All authors were involved in data analyses, contributed critically to the drafts, and gave final approval for publication.

Abstract

The parallel evolution of similar ecotypes in response to comparable environmental conditions is believed to reveal the importance of divergent selection in phenotypic diversifying processes. Systems characterized by the presence of multiple replicate populations expressing resource polymorphism thus provide an ideal opportunity to address the occurrence and factors affecting the parallel evolution of ecotypes. Previous studies have shown that brook charr (*Salvelinus fontinalis*) exhibit resource polymorphism in some Canadian Shield lakes, where a littoral ecotype feeds mainly on zoobenthos and a pelagic ecotype feeds mostly on zooplankton. Using morphological traits and geometric morphometric analyses on 18 native brook charr populations, we explicitly tested that (i) brook charr ecotypes show parallel evolution across populations (i.e., the same morphological traits discriminate ecotypes among lakes) and (ii) interspecific competition decreases the magnitude of morphological differentiation between ecotypes, if any (because brook charr experience some level of competitive exclusion from the littoral habitat in the presence of creek chub or white sucker). We observed a low level of parallel evolution, where the littoral ecotype was overall stouter with longer fins and smaller eyes than the pelagic ecotype. Interspecific competition had no clear impacts on the magnitude of morphological differentiation. We also observed that inter-lake morphological differences are more important than intra-lake habitat specialization (i.e., morphological differences between ecotypes), suggesting an important effect of local environmental factors on population morphology. Early-stage diversification as well as phenotypic plasticity and morphological integration could explain why resource polymorphism is still subtle in brook charr populations.

Keywords: divergent selection, geometric morphometrics, phenotypic plasticity, morphological differentiation, resource polymorphism.

Introduction

Understanding the mechanisms underlying ecological speciation is an iconic objective of ecology, and studying incipient species is among the best approaches to shed light on early diversifying processes. Resource polymorphism, i.e., when discrete phenotypes of the same population show differential niche use, is believed to represent an important intermediate stage in sympatric speciation (Smith and Skúlason 1996; Schluter 2000; Skúlason et al. 2019). It is generally accepted that the first requirement for resource polymorphism to occur is the presence of an ecological contrast (i.e., discrete resources or habitat; Hendry 2009). However, these ecological contrasts can lead to divergent selection only when they impose trade-offs among traits, constraining within-individual niche width during niche expansion at the benefit of between-individual variation (Bolnick et al. 2003). This mechanism explains why a generalist exploiting two resources would not perform as well as the specialists and would be selected against (Bolnick et al. 2003). Another factor promoting resource polymorphism would be the release from interspecific competition (Smith and Skúlason 1996; Martin and Pfennig 2010), as it generally contributes to increase access to new underexploited resources, i.e., ecological opportunities (Martin and Pfennig 2010).

The nature of resource polymorphism involves a close connection between resource-use efficiencies and traits defining ecotypes (e.g., morphological, behavioural, or physiological variations). In this context, similar selective pressures should impose common evolutionary responses among populations (Adams 2010). Therefore, the parallel evolution of ecotypes, which occurs when similar discrete phenotypes evolve in multiple populations adapting to comparable habitats, should be strong evidence for the importance of divergent selection in resource polymorphism processes (Schluter 2000). Parallel evolution has been observed in many systems exhibiting resource polymorphism, such as amphibians (e.g., Rice et al. 2009; Adams 2010), birds (e.g., Smith 1997; Hugall and Stuart-Fox 2012), and fish from post glacial lakes (e.g., Sigursteinsdóttir and Kristjánsson 2005; Østbye et al. 2006; Jacobs et al. 2020). However, a recent meta-analysis revealed high variability in the level of parallelism between ecotypes of replicate

populations (Oke et al. 2017), highlighting the lack of knowledge on factors driving the level of phenotypic parallelism in this situation.

In northern lakes, the most common form of parallel evolution is indisputably the adaptation of discrete ecotypes to either littoral or pelagic resources (Robinson and Wilson 1994; Smith and Skúlason 1996). These ecotypes often present morphological adaptations associated with swimming demands and feeding efficiency when using littoral or pelagic habitats (Robinson and Parsons 2002). This common pattern of resource specialization in addition to the functional expectations related to habitat divergence suggests that these evolutionary responses are indeed selective (Robinson and Parsons 2002). Brook charr, *Salvelinus fontinalis*, exhibit a subtle resource polymorphism in some Canadian Shield lakes (Bourke et al. 1997; Dynes et al. 1999; Proulx and Magnan 2004; Bertrand et al. 2008; Chapter III). The littoral ecotype is generally stouter and has longer pectoral fins, whereas the pelagic ecotype is more fusiform and has shorter pectoral fins. It was also observed that some morphological traits and feeding behaviours are transmitted from parents to the next generation (Proulx and Magnan 2004; Sacotte and Magnan 2006). Morphological differentiation associated with this resource polymorphism is also functionally related to the swimming performance and energetics of the two ecotypes (Proulx and Magnan 2002; Peres-Neto and Magnan 2004; Rouleau et al. 2010).

Bait fishers introduced creek chub (*Semotilus atromaculatus*) and white sucker (*Catostomus commersonii*) to many of these lakes over the last century. These introduced species compete for food with brook charr in the littoral zone (Magnan 1988; Tremblay and Magnan 1991; Lacasse and Magnan 1992), decreasing its relative abundance and biomass by 30% to 70% (Magnan 1988; Lachance and Magnan 1990; Magnan et al. 2005). In this system, the level of interspecific competition varies from none (allopatric brook charr lakes), to intermediate (brook charr with creek chub lakes), to high (brook charr with creek chub and white sucker lakes) (Magnan 1988; Bourke et al. 1999). This gradient in the intensity of interspecific competition provides a good opportunity to address how it shapes the morphological diversification between ecotypes and among the fish communities (i.e., ecological release).

Despite much evidence for resource polymorphism in brook charr at the lake level (Bourke et al. 1997; Dynes et al. 1999; Proulx and Magnan 2004; Bertrand et al. 2008; Chapter III), the parallel evolution of these ecotypes has not yet been explicitly tested. The goals of this study were thus to test that (i) brook charr ecotypes show parallel evolution across populations (i.e., whether the same morphological traits discriminate ecotypes among lakes), and (ii) interspecific competition decreases the magnitude of morphological differentiation between ecotypes, if indeed any is observed (i.e., because brook charr experience some level of competitive exclusion from the littoral habitat in the presence of creek chub or white sucker). To this end, we sampled brook charr from 18 Canadian Shield lakes along the gradient of interspecific competition described above to determine if divergent selection forces are similar among brook charr populations. We used both morphologic traits and body shape to characterize morphological differentiation between littoral and pelagic brook charr.

Materials and Methods

Study area and fish sampling

The study was conducted in Mastigouche (46° 40′ N, 73° 30′ W) and Saint-Maurice (47° 05′ N, 73° 15′ W) wildlife reserves, and in La Mauricie National Park (46° 45′ N, 73° 08′ W), Québec, Canada, from June to August 2012, 2013, and 2014. We sampled seven lakes containing only brook charr (BC), six with brook charr and creek chub (BC+CC), and five with brook charr, creek chub, and white sucker (BC+CC+WS; Table 1). Other fish species were found in BC+CC and BC+CC+WS lakes (Online Resource Table 1), but the main competitors of brook charr in this system are creek chub and white sucker (Magnan 1988, Magnan et al. 2005). The average biomass per trap (BPUE) of the two main competitors in the littoral habitat was 2.8 kg/trap for BC+CC lakes and 6.0 kg/trap for BC+CC+WS lakes. The average creek chub BPUE in the littoral habitat was over three times higher in BC+CC lakes (2.8 kg/trap) compared to BC+CC+WS lakes (0.9 kg/trap). Brook charr BPUE was similar between BC and BC+CC lake (~1.2 kg/trap) but 25% lower in BC+CC+WS lakes (~0.9 kg/trap). Fish were caught

in both the littoral (< 3 m) and pelagic (> 4 m) zones of lakes. The 3 m depth represents the limit of the littoral zone in this system, as defined by Wetzel and Likens (2000): "the zone containing a substantial number of microhabitats associated with surfaces of submersed macrophytes, with particulate detritus, and with the sediments." We arbitrarily considered the beginning of the pelagic zone to be 4 m of depth to prevent any overlap with the littoral zone. Fish were caught with four trap nets (Alaska type; opening 1.0 m × 1.8 m, equipped with two 1 m × 15 m wings; Fipec Industries, Gaspé, Québec, Canada). The traps were located randomly in both the littoral and pelagic zones and fished for four (2013-2014) or five (2012) consecutive days from 18:00 to 06:00 to capture brook charr at the time they are the more active in their feeding habitat (based on previous studies in this system; Bourke et al. 1996; Bertolo et al. 2011; Goyer et al. 2014). The mean depths (± SD) of trap nets were 2.1 ± 0.8 m and 5.2 ± 1.0 m in the littoral and pelagic zones, respectively. We randomly selected 20 adult brook charr from both zones of each lake with total lengths between 19 cm and 30 cm to minimize ontogenetic morphologic changes and to ensure the selection adult individuals. Each brook charr was measured (total length, jaw width, and body width; mm), weighed (g), sexed, and photographed on the left side (on a Styrofoam plate carved to allow individuals to lie flat in a lateral position) against a reference scale for post-processing standardization. Jaw width and body width were measured in the field with an electronic caliper. Optical distortions (which are higher in the border area of photographs) were minimized by photographing specimens from a distance of 50-70 cm depending on brook charr size.

Morphometric measurements

A total of 25 landmarks were placed on each fish picture, of which 15 were used for body shape analysis, 21 for trait analysis, and four for "unbending" specimens (Fig. 1). We used tpsDig 2 to set landmarks (Rohlf 2012a), and all landmarks were positioned by the same person. We used tpsUtil to create .tps files from pictures and randomly ordered specimens before positioning the landmarks (Rohlf 2012b). We also used tpsUtil to "unbend" specimens using the landmark of the tip of the snout, two landmarks along the lateral line, and one at the end of the caudal pedunculus (Dermond et al. 2019).

Morphological trait analysis

A total of 17 morphological traits related to feeding and swimming behaviours were used in this analysis (Fig. 1). As proposed by Oke et al. (2017), we first quantified the percentage of variance explained by the ecotype designation and its effect size. To do so, all morphological traits and total length were log transformed ($\log_e$ x) and standardized. We then removed the effect of body size and sex for each lake population independently using linear modelling techniques (Fleming et al. 1994; Reist 1986). The procedure followed the general recommendation of "common within-group" regression slope (Reist 1986; Fleming et al 1994). Once corrected, we used simple univariate ANOVAs to assess the effect size and the percentage of variance that could be explained by the ecotype. We also estimated the effect size and semi-partial R^2 of ecotype designation across populations using mixed models with body size and sex as covariables and lake as the random effect for each of the 17 traits. All analyses were carried out with the R software (R Development Core Team, 2014). We used the "nlme" package (3.1-137) for all mixed-models and the "r2beta" function (r2glmm package; v. 0.1.2) to compute semi-partial R^2.

Second, we addressed the phenotypic variation between ecotypes using all 17 traits with discriminant analysis. Do to so, we removed the effect of body size, sex, and lake using linear mixed modelling techniques. The procedure followed the general recommendation of the "common within-group" regression slope (Reist, 1986; Fleming et al. 1994) but was extended to multiple sources of variation (groups and covariates) for nested data (i.e., using mixed models; Pépino et al. 2018). Performing accurate adjustments for size as well as correcting for undesirable and noisy variables (e.g., sex and lake variables) was important to do before group discrimination because of error propagation in further analyses (Reist 1986; Pépino et al. 2018). Zuur et al. (2007) recommend verifying the linear relationships among variables before conducting discriminant function analyses and suggest removing one variable when the correlation between two is 0.9 or higher. Relationships among adjusted morphological traits were approximately linear, with positive correlations that never exceeded a Pearson correlation of 0.77.

We performed linear discriminant function analysis (LDA) using the 17 adjusted morphological traits as descriptors and the Ecotype × Fish community as the group variable. We defined six distinct groups as the littoral and pelagic ecotypes of the three fish communities (BC, BC+CC, and BC+CC+WS). Because LDA is sensitive to outliers, 16 out of 700 individuals were removed before adjustment and LDA. Following the general recommendations of Borcard et al. (2011), we verified the condition of multivariate homogeneity of within-group covariance (permutation test; n = 999, $F_{5, 678}$ = 1.39, p = 0.23). A Jackknife-based classification (i.e., leave-one-out cross-validation) was applied to estimate the accuracy of the discrimination between the six groups (Lance et al. 2000; Olden et al. 2002). Finally, we calculated the correlation between the original descriptors (adjusted morphological traits) and the canonical scores (also called canonical correlation coefficients; Zuur et al., 2007) to better interpret the relationship between group discrimination and morphological variation.

Geometric morphometric analysis

We also used linear discriminant function analysis to determine the amount of shape variation attributable to sex, ecotype, and fish community. We first performed a Procrustes superimposition on the 15 landmarks (Fig. 1) to correct for differences in size, position, and orientation of fish in pictures (Rohlf and Slice 1990, Rohlf and Marcus 1993). The LDA was done separately on male and female brook charr because this species has sexual dimorphism (Proulx and Magnan 2004). We used the 15 body landmarks as the response variables and the Ecotype × Fish community as the group variable. We defined six distinct groups as the littoral and pelagic ecotypes of the three fish communities (BC, BC+CC, and BC+CC+WS). Because LDA is sensitive to outliers, 12 out of 700 individuals were removed before adjustment and LDA, resulting in 342 males and 346 females available for analysis. A Jackknife-based classification (i.e., leave-one-out cross-validation) was applied to estimate the accuracy of the discrimination between the six groups (Lance et al. 2000; Olden et al. 2002). We represented the extreme of each axis to better interpret the relationship between group discrimination and body shape variations.

We quantified the relative amount of shape variation attributable to sex, ecotype, and fish community using distance-based MANOVA. This method is suitable for high-dimensional data such as morphometric data (Collyer et al. 2015; Collyer and Adams 2018). In this analysis, we included lake as the random effect and centroid size (calculated during the Procrustes superimposition) to consider possible ontogenic morphological differences among individuals.

Finally, we conducted phenotypic trajectory analyses (PTA; Collyer and Adams 2013), also called phenotypic change vector analyses (PCVA) in the specific case of only two points along the trajectory (Oke et al. 2017), on the shape coordinates to assess whether the trajectory of phenotypic change between littoral and pelagic ecotypes differs among lakes. This procedure connects littoral to pelagic ecotypes of each lake in multidimensional shape space, and the resulting trajectories are defined by two attributes – vector length (or magnitude; d) and direction. Sampling distributions from 1000 random permutations were used in pairwise comparisons to test magnitude differences (i.e., difference in trajectory path lengths) and trajectory correlations (i.e., angular differences between trajectory directions) among lakes. No significant differences in either attribute under the null hypothesis (i.e., $\Delta d = 0$ or $\theta = 0$ for differences in vector length and direction, respectively) were rejected if the p-value of the observed attribute difference was less than $\alpha = 0.05$. PCVA were performed by first fitting the model with the "procD.lm" function and then using this model in the function "trajectory.analysis" with lakes as the group argument and ecotype as the trajectory points argument.

All analyses were performed in the R environment (R Development Core Team, 2014). We used the "MASS" package (v. 7.3-51.4; Venables and Ripley 2002) for LDA and the "candisc" package (v. 0.8-0; Friendly and Fox 2017) for calculating the canonical correlation coefficients. Geometric morphometric analyses were performed using the "geomorph" package (v. 3.1.3; Adams et al. 2019).

Results

Analysis of morphological traits

Mixed model trait analyses revealed that the percentages of variance explained by ecotype ranged from 0.4% to 16% (semi-partial R^2) for the 17 traits selected in this study and was 4% on average (Fig. 2). The maximum variance explained by ANOVAs (R^2) was 48% for the caudal peduncle height but remained low on average (4.9%). Most traits showed divergent effect sizes among lakes (i.e., both positive and negative effects of the ecotype for the same trait in ANOVAs; Fig. 2). The mixed model approach revealed that differences between ecotypes were significant for 9 of the 17 traits, ($p < 0.05$; Fig. 2). Among these traits, jaw width, body width, pectoral fin, caudal peduncle height, head height, dorsal fin base, pelvic fin height, and body height were longer for the littoral ecotype while eye was wider for the pelagic ecotype.

Linear discriminant function analysis showed that the first two discriminant axes represented more than 90% of the total variation (Fig. 3a). Wilk's λ statistic indicated a significant group effect ($\lambda = 0.43$, $p < .001$). The first axis mostly discriminated the three fish communities and accounted for 76.4% of the total variation, while the second axis partially discriminated allopatric vs. sympatric populations (13.7 % of the total variation; Fig. 3a). Both ecotypes showed large overlaps within the fish community, with a small trend to shift toward negative values along the first axis for the littoral ecotype. Canonical correlation coefficients of traits revealed that the first axis is positively related to body condition (i.e., higher body height BH, body width BW, dorsal fin base DB, and peduncle height PS) and negatively related to eye size (EW) and head morphology (higher head length HL, upper jaw SJ, and snout to posterior cranium SC). The second axis, which explained only 13.7% of the total variation, is negatively related to ventral fins (pectoral fin PC, pelvic fin PH, and anal fin AH), mouth width (MW), and eye size (EW; Fig. 3b). Overall, brook charr from BC+CC+WS lakes were stouter compared to those from BC+CC lakes, which were more fusiform (i.e., longer head and a slender body) with larger eyes (EW; Fig. 3b). The morphological traits of brook charr from BC lakes fell between the other two fish communities but had slightly higher positive values along the

second axis, with shorter pectoral (PC), pelvic (PH), and anal (AH) fins and reduced eye size (EW) and mouth width (MW; Fig. 3a). Jackknife-based classification varied from 22.8% (littoral ecotype from BC+CC+WS lakes) to 51.3% (pelagic ecotype from BC+CC+WS lakes) and was overall 36% (a random classification would have been ~16.7% for each group; Table 2). Misclassifications were higher for individuals from the same fish community (Table 2).

Geometric morphometric analysis

For males, linear discriminant function analysis showed that the first two discriminant axes represent around 75% of the total variation (Fig. 4a). Wilk's λ statistic indicated a significant group effect ($\lambda = 0.33$, $p < 0.001$). The first axis mostly discriminated BC and BC+CC+WS from BC+CC, while the second axis discriminated fish communities according to the interspecific competition gradient – from BC lakes (negative PC2 values) to BC+CC+WS lakes (positive PC2 values) – but with a large overlap among fish communities. Both ecotypes showed a large overlap within fish communities, with a weak trend to more positive values along PC1 for the pelagic ecotype. The main body shape differences were found along the first axis, where individuals ranged from deeper and compact bodies (negative PC1 values) to more extended, fusiform, and slim bodies (positive PC1 values). Body shape differences were weak along the second axis: negative PC2 values tended to be associated with individuals having shorter heads, dorsal fins shifted to the rear, and pelvic fins shifted toward the front. Jackknife-based classification varied from 26.2% (littoral ecotype from BC+CC+WS lakes) to 51.3% (pelagic ecotype from BC+CC+WS lakes) and was overall 33.6% (a random classification would have been 16.7% for each group; Table 2). Misclassifications were higher for individuals that belonged to the same fish community but were classified as the opposite ecotype (Table 2).

Linear discriminant function analysis with females showed that the first two discriminant axes represented ~77% of the total variation (Fig. 4b). Wilk's λ statistic indicated a significant group effect ($\lambda = 0.35$, $p < 0.001$). The first axis mostly discriminated BC and

BC+CC+WS from BC+CC, while the second axis discriminated BC and BC+CC from BC+CC+WS (Fig. 4b). No discrimination of the two ecotypes was apparent. The main body shape differences ranged from deeper individuals (negative PC1 values or positive PC2 values) to slim individuals (positive PC1 values or negative PC2 values). Jackknife-based classification varied from 18.5% (littoral ecotype from BC+CC+WS lakes) to 42.3% (pelagic ecotype from BC+CC+WS lakes) and was overall 32.4% (a random classification would have been 16.7% for each group; Table 2). Misclassifications were higher for individuals that belonged to the same fish community but were classified as the opposite ecotype (Table 2).

The PCVA (phenotypic change vector analyses) revealed that shape variation was more important among lakes than between ecotypes (Fig. 5a). Indeed, the median inter-lake distance in multidimensional shape space was about two times higher than the median of the magnitude (i.e., vector length between littoral and pelagic ecotypes). PC1 explained ~44% of the inter-group variation, ranging from stouter (negative PC1 values) to slimmer individuals (positive PC1 values; Fig. 5b). Along this axis, the three landmarks with the greatest deformation were associated with body width (i.e., two landmarks at the base of the dorsal fin and one landmark at the insertion of the pelvic fin) and, to a lesser extend, with head length (i.e., longer head length for positive PC1 values). The PC2 explained ~20% of the inter-group variation. Along this axis, the elongation of the head from negative to positive PC2 values was probably the most relevant shape variation. For differences in magnitude, 16 out of 153 pairwise comparisons were statistically significant ($p < 0.05$; Table 3). Three lakes were involved in these 16 significant pairwise comparisons and were associated with the first three lakes with the highest magnitudes: lakes R, W, and O, with nine, six, and one significant pairwise comparisons, respectively. Lakes R and O were BC+CC+WS lakes whereas lake W was a BB+BC lake. Although there was a general trend to have smaller magnitude trajectories for allopatric lakes, no significant differences were found among fish communities (ANOVA: $F_{2,15} = 1.374$; $p = 0.283$). For differences in direction, 13 out of 153 pairwise comparisons were statistically significant ($p < 0.05$; Table 4). We did not detect any pattern in lakes involved in these significant differences. Overall, most of the pairwise comparisons were

not statistically significant ($p > 0.05$) for either magnitude or direction attributes of the trajectories, suggesting parallel evolution or a lack of statistical power to detect differences. Fifteen out of 18 trajectories started from more negative PC1 values, suggesting that the littoral ecotype was stouter than the pelagic one within a given lake. Nevertheless, the magnitude was weak and, although not statistically significant, the angle difference averaged 87° (in the multidimensional shape space), suggesting orthogonal trajectories. Finally, we did not observe any evidence of an effect of fish communities on trajectories: all fish communities seemed well mixed except for three out of six BC+CC lakes that were located at the positive extremity of PC1 (Fig. 5a).

The MANOVA results corroborated results obtained with the PCVA and showed that the among-lake body shape variation represented most of the explained variation compared to sex, ecotype, or fish community (Table 5). Overall, body shape analyses revealed that brook charr from BC+CC lakes are slimmer than individuals from either BC or BC+CC+WS lakes.

Discussion

Overall, the results of this study-based on both morphological trait (LDA) and body shape (PCVA) analyses – revealed that the among-lake morphological differences are more important than intra-lake habitat specialization (i.e., morphological differences between ecotypes). However, the results show some degree of parallel evolution in brook charr ecotypes. Although the variation of each trait explained by the ecotype are weak, the differences between littoral and pelagic brook charr were significant for 9 of the 17 traits considered. The results reveal that littoral brook charr are stouter (higher jaw width, body width and height, head height, caudal peduncle height), has longer/larger fins (higher dorsal fin base and length, pelvic fin height), and smaller eyes than the pelagic ecotype. The trajectory analysis based on shape coordinates also support that the littoral ecotype is stouter than the pelagic one in 15 of the 18 study lakes. Finally, we did not find support for the hypothesis that interspecific competition would decrease the morphological differentiation between ecotypes by restricting individuals to resources in

the open-water zone. Instead, our results suggest that most of the morphological variation comes from local adaptations (i.e., at the lake level). Furthermore, brook charr from BC lakes were more similar to those from BC+CC+WS lakes than to those from BC+CC lakes, which were slimmer compared to brook charr from the other two fish communities, suggesting a lower body condition in these populations.

Extent of parallel evolution in brook charr ecotypes

Different factors could explain the low percentage of variation in morphological traits and body shapes explained by the ecotype term and why many traits showed divergent effect size among lakes (and thus low parallel evolution). First, considering the sample size of fish considered in our analyses and the fact that the resource polymorphism in brook charr is subtle (Dynes et al. 1999; Peres-Neto and Magnan 2004; Proulx and Magnan 2004; Bertrand et al. 2008), there could have been a lack of statistical power to detect small differences between littoral and pelagic ecotypes. Indeed, the amount of variation in brook charr traits explained by the ecotype term is low compared to those reported by Oke et al. (2017) in their meta-analysis, and thus would have required a higher sample size to detect significant differences. In support of this, our mixed model analysis to estimate the effect size of the ecotype term across populations with body size and sex as covariable and were significant in 9 out of the 17 traits considered. Mixed models use information from the data much more efficiently than ANOVA by shrinking estimates to the mean in a global analysis. Second, as suggested by Oke et al. (2017), traits used in morphological studies are often measured for convenience (or routinely), not because they are expected to experience parallel divergent selection. A general lack of clear predictions on whether individual traits were expected to show parallelism precluded the ability to remove traits that might be unrelated to divergence or not expected to be under selection. In this context, one should benefit from careful consideration of whether each individual trait has a relevant link to fitness during divergence and should be included in estimates of the extent of parallelism (Oke et al. 2017). Third, and in line with the latter point, some traits are phenotypically flexible and possibly not under strong selection. For example, in a common garden experiment, Proulx and Magnan (2004) showed that some characters remained

unchanged during a four-month shift of brook charr from their initial to the opposite habitat while some others exhibited a complete reversal, in agreement with the predictions of functional morphology. This result indicates that some characters are under pure environmental control and not fixed after a fish has adopted a given strategy (Proulx and Magnan 2004). In addition, brook charr are known to present a low degree of morphological integration, which allows most traits to respond independently to environmental drivers (Peres-Neto and Magnan 2004). These characteristics should allow a phenotypic response to small differences in habitats across lakes, leading to non-parallel phenotypic divergence between ecotypes. Our results suggest that each lake has a set of distinct environmental characteristics forging local adaptation at a population level.

The morphological traits and body shapes characterizing both brook charr ecotypes reported above meet the functional expectations associated with foraging in their respective habitats. Based on these predictions, littoral ecotypes should be characterized by deeper bodies, larger heads, longer paired fins (pectoral or pelvic), and deeper but compressed caudal peduncles, while pelagic ecotypes should have slender elongated bodies, smaller paired fins, and shallower but wider caudal peduncles (Robinson and Parson 2002). These traits are thus more likely to show parallel evolution among populations because they are associated with the swimming demands of each habitat. These variations between ecotypes specialized to similar habitats were also observed in an arctic charr population, where a small benthic ecotype from different lakes showed similarities but also subtle morphological differences that could indicate local specialized adaptations (Sigursteinsdóttir and Kristjánsson 2005).

Effect of interspecific competition on parallel evolution

The morphological differences between ecotypes were of the same magnitude along the gradient of interspecific competition, which contradicts our second hypothesis. In the same way that chapter IV revealed that when facing interspecific competition from creek chub and white sucker, both littoral and pelagic brook charr incorporated more pelagic prey into their diet but maintained the amplitude of their differences in resource use.

The feeding mode of brook charr (particulate feeder) differs from that of white sucker (bottom suction feeder), and even though creek chub are particulate feeders, they are gape-limited in prey size compared to brook charr. Chapter III suggested that these differences could leave a portion of the littoral niche available for brook charr in lakes with competitors, explaining why some individuals still use this habitat in sympatry. Of course, the carrying capacity of the littoral zone for brook charr is lower in sympatric populations, explaining why their abundance and biomass are lower in these communities (Magnan 1988; Lachance and Magnan 1990; Magnan et al. 2005). These results suggest that the level of intraspecific competition among brook charr remains comparable in allopatric and sympatric with creek chub and white sucker populations (i.e., between BC and BC+CC+WS), explaining why the two ecotypes are maintained and that their resource specialization as well as their morphology (in the present study) remain comparable in both of these two fish communities. Interestingly, body shape analysis revealed that brook charr are slimmer in BC+CC lakes suggesting lower body condition compared to other fish communities. The brook charr biomass (BPUE) were not significantly different between BC and BC+CC lakes and significantly lower in BC+CC+WS lakes. In this system, creek chub BPUE is over three times higher in BC+CC compared to BC+CC+WS lakes and brook charr incorporates more pelagic prey into their diet along this gradient of increasing interspecific competition (Chapter III). These results suggest that intra-specific competition experienced by brook charr is higher in BC+CC lakes because inter-specific competition by creek chub reduces the resources availability but not brook charr biomass (BPUE).

Conclusion

This study suggests that the magnitude of morphological differentiation between ecotypes is still low in lacustrine brook charr populations of the Canadian Shield, even though some morphological characteristics seem to show a parallel evolution in most replicate populations, with littoral brook charr being stouter with longer fins and smaller eyes than pelagic ones. This study also emphasized that morphological differences between ecotypes are not consistent across populations for most traits and that among-lake

morphological differences are more important than morphological differences between ecotypes. This observation echoes a recent meta-analysis addressing the extent of parallel evolution among polymorphic fish populations that clearly showed that the extent of parallelism between littoral and pelagic ecotypes is highly variable across populations of the same species (Oke et al. 2017). Finally, these lakes were formed following the retreat of the North America ice sheets about 15,000 years ago and were subsequently colonized by brook charr from glacial refugia (Angers and Bernatchez 1998). Among the factors that could influence morphological differentiation between ecotypes, interspecific competition by recently introduced fish species does not seem to have a high impact on the morphological trajectories between ecotypes. Thus, the low level of parallel evolution observed in this study between littoral and pelagic ecotypes could be the result to an early stage of diversification as well as the high level of plasticity and morphological integration of brook charr. This make such systems of particular interest for understanding processes involved in parallel evolution.

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References

Adams DC (2010) Parallel evolution of character displacement driven by competitive selection in terrestrial salamanders. BMC Evolutionary Biology 72: 1-10. doi: 10.1186/1471-2148-10-72

Adams DC, Collyer ML, Kaliontzopoulou A (2019) Geomorph: Software for geometric morphometric analyses. R package version 3.1.0. https://cran.r-project.org/package=geomorph.

Angers B, Bernatchez L (1998). Combined use of SMM and non-SMM methods to infer fine structure and evolutionary history of closely related brook charr (*Salvelinus fontinalis*, Salmonidae) populations from microsatellites. Molecular Biology and Evolution 15, 143-159.

Bertolo A, Pépino M, Adams J, Magnan P (2011) Behavioural thermoregulatory tactics in lacustrine brook charr, *Salvelinus fontinalis*. PLoS One 6:e18603. doi: 10.1371/journal.pone.0018603

Bertrand M, Marcogliese DJ, Magnan P (2008) Trophic polymorphism in brook charr revealed by diet, parasites and morphometrics. Journal of Fish Biology 72: 555-572. doi: 10.1111/j.1095-8649.2007.01720.x

Bolnick DI, Svanbäck R, Fordyce JA, Yang LH, Davis JM, Hulsey CD, Forister, ML (2003) The ecology of individuals: incidence and implications of individual specialization. The American Naturalist 161: 1-28. doi: 10.2307/3078879

Borcard D, Gillet F, Legendre P (2011). Numerical ecology with R. New York, NY: Springer.

Bourke P, Magnan P, Rodriguez MA (1996) Diel locomotor activity of brook charr, as determined by radiotelemetry. Journal of Fish Biology 49: 1174-1185. doi: 10.1006/jfbi.1996.0245

Bourke P, Magnan P, Rodriguez MA (1997) Individual variations in habitat use and morphology in brook charr. Journal of Fish Biology 51: 783-794. doi: 10.1006/jfbi.1997.0481

Bourke P, Magnan P, Rodriguez MA (1999) Phenotypic responses of lacustrine brook charr in relation to the intensity of interspecific competition. Evolutionary Ecology 13: 19-31.

Collyer ML, Adams DC (2013) Phenotypic trajectory analysis: comparison of shape change patterns in evolution and ecology. Hystrix, the Italian Journal of Mammalogy 24: 75-83. doi:10.4404/hystrix-24.1-6298

Collyer ML, Adams DC (2018) RRPP: An R package for fitting linear models to high-dimensional data using residual randomization. Methods in Ecology and Evolution 78: 43-49. doi: 10.1111/2041-210X.13029

Collyer ML, Sekora DJ, Adams DC (2015) A method for analysis of phenotypic change for phenotypes described by high-dimensional data. Heredity 115: 357-365. doi: 10.1038/hdy.2014.75

Dermond P, Sperlich N, Brodersen J (2019) Heritable morphological differentiation in salmonids from two distinct stream types. Journal of Fish Biology 95: 1215-1222. doi: 10.1111/jfb.14121

Dynes J, Magnan P, Bernatchez L, Rodriguez MA (1999) Genetic and morphological variation between two forms of lacustrine brook charr. Journal of Fish Biology 54: 955-972.

Fleming IA, Jonsson B, Gross MR (1994). Phenotypic divergence of sea-ranched, farmed, and wild salmon. Canadian Journal of Fisheries and Aquatic Sciences 51: 2808-2824.

Friendly M, Fox J (2017). candisc: Visualizing Generalized Canonical Discriminant and Canonical Correlation Analysis. R package version 0.8-0. https://CRAN.R-project.org/package=candisc.

Goyer K, Bertolo A, Pépino M, Magnan P (2014) Effects of lake warming on behavioural thermoregulatory tactics in a cold-water stenothermic fish. PLoS One 9:e92514. doi: 10.1371/journal.pone.0092514

Hendry A (2009) Ecological speciation! Or the lack thereof? Canadian Journal of Fisheries and Aquatic Sciences 66: 1383-1398. doi: 10.1139/F09-074

Hugall AF, Stuart-Fox D (2012) Accelerated speciation in colour-polymorphic birds. Nature 485: 631-634. doi: 10.1038/nature11050

Jacobs A, Carruthers M, Yurchenko A, Gordeeva NV, Alekseyev SS, Hooker O, Leong JS, Minkley DR, Rondeau EB, Koop BF, Adams CE, Elmer KR (2020) Parallelism in eco-morphology and gene expression despite variable evolutionary and genomic backgrounds in a Holarctic fish. PLoS Genetics 16: e1008658. doi: 10.1371/journal.pgen.1008658

Lacasse S, Magnan P (1992) Biotic and abiotic determinants of the diet of brook trout, *Salvelinus fontinalis*, in lakes of the Laurentian Shield. Canadian Journal of Fisheries and Aquatic Sciences 49: 1001-1009.

Lachance S, Magnan P (1990) Performance of domestic, hybrid, and wild strains of brook trout, *Salvelinus fontinalis*, after stocking: the impact of intra- and interspecific competition. Canadian Journal of Fisheries and Aquatic Sciences 47: 2278-2284. doi: 10.1139/f90-253

Lance RF, Kennedy ML, Leberg PL (2000). Classification bias in discriminant function analyses used to evaluate putatively different taxa. Journal of Mammalogy 81: 245-249.

Magnan P (1988) Interaction between Brook Charr, *Salvelinus fontinalis*, and nonsalmonid species: Ecological shift, morphological shift, and their impact on zooplankton communities. Canadian Journal of Fisheries and Aquatic Sciences 45: 999-1009.

Magnan P, Proulx R, Plante M (2005) Integrating the effects of fish exploitation and interspecific competition into current life history theories: an example with lacustrine brook trout (*Salvelinus fontinalis*) populations. Canadian Journal of Fisheries and Aquatic Sciences 62: 747-757. doi: 10.1139/F05-041.

Martin RA, Pfennig DW (2010) Field and experimental evidence that competition and ecological opportunity promote resource polymorphism. Biological Journal of the Linnean Society 100: 73-88. doi: 10.1111/j.1095-8312.2010.01380.x

Oke KB, Rolshausen G, LeBlond C, Hendry AP (2017) How parallel is parallel evolution? A comparative analysis in fishes. The American Naturalist 190: 1-16 doi: 10.1086/691989

Olden JD, Jackson DA, Peres-Neto PR (2002). Predictive models of fish species distributions: A note on proper validation and chance predictions. Transactions of the American Fisheries Society 131: 329-336.

Østbye K, Amundsen PA, Bernatchez L, Klemetsen A, Knudsen R, Kristoffersen R, Næsje TF, Hindar K (2006) Parallel evolution of ecomorphological traits in the European whitefish *Coregonus lavaretus* (L.) species complex during postglacial times. Molecular Ecology 15: 3983-4001. doi: 10.1111/j.1365-294X.2006.03062.x

Pépino M, Magnan P, Proulx R (2018) Field evidence for a rapid adaptive plastic response in morphology and growth of littoral and pelagic brook charr: A reciprocal transplant experiment. Functional Ecology 32: 161-170. doi: 10.1111/1365-2435.12929

Peres-Neto PR, Magnan P (2004) The influence of swimming demand on phenotypic plasticity and morphological integration: a comparison of two polymorphic charr species. Oecologia 140: 36-45. doi: 10.1007/s00442-004-1562-y

Pfennig DW, Wund MA, Snell-Rood EC, Cruickshank T, Schlichting CD, Moczek AP (2010) Phenotypic plasticity's impacts on diversification and speciation. Trends in Ecology and Evolution 25: 459-467. doi: 10.1016/j.tree.2010.05.006

Proulx R, Magnan P (2002) Physiological performance of two forms of lacustrine brook charr, *Salvelinus fontinalis*, in the open-water habitat. Environmental Biology of Fishes 64: 127-136.

Proulx R, Magnan P (2004) Contribution of phenotypic plasticity and heredity to the trophic polymorphism of lacustrine brook charr (*Salvelinus fontinalis* M.). Evolutionary Ecology Research 6: 503-522.

R Development Core Team (2014). R: A language and environment for statistical computing. Vienna, Austria: R Foundation for Statistical Computing. Retrieved from https://www.r-project.org.

Reist JD (1986). An empirical evaluation of coefficients used in residual and allometric adjustment of size covariation. Canadian Journal of Zoology 64: 1363-1368.

Rice AM, Leichty AR, Pfennig DW (2009) Parallel evolution and ecological selection: replicated character displacement in spadefoot toads. Proceedings of the Royal Society B: Biological Sciences 276: 4189-4196. doi: 10.1098/rspb.2009.1337

Robinson BW, Parsons KJ (2002) Changing times, spaces, and faces: tests and implications of adaptive morphological plasticity in the fishes of northern postglacial lakes. Canadian Journal of Fisheries and Aquatic Sciences 59: 1819-1833. doi: 10.1139/f02-144

Robinson BW, Wilson DS (1994) Character release and displacement in fishes: a neglected literature. The American Naturalist 144: 596-627.

Rohlf FJ (2012a) tpsDig2, Version 2.16 [computer program]. Department of Ecology and Evolution. State University of New York, Stony Brook https://life.bio.sunysb.edu/morph/

Rohlf FJ (2012b) tpsUtil, Version 1.57 [computer program]. Department of Ecology and Evolution. State University of New York, Stony Brook https://life.bio.sunysb.edu/morph/.

Rohlf FJ, Slice DE (1990) Extensions of the Procrustes method for the optimal superimposition of landmarks. Systematic Zoology. 39: 40-59.

Rohlf FJ, Marcus L (1993) A revolution in morphometrics. Trends in Ecology and Evolution 8: 129-132 doi: 10.1016/0169-5347(93)90024-J

Rouleau S, Glémet H, Magnan P (2010) Effects of morphology on swimming performance in wild and laboratory crosses of brook trout ecotypes. Functional Ecology 24: 310-321. doi: 10.1111/j.1365-2435.2009.01636.x

Sacotte S, Magnan P (2006) Inherited differences in foraging behaviour in the offspring of two forms of lacustrine brook charr. Evolutionary Ecology Research 8: 843-857.

Schluter D (2000) The ecology of adaptive radiation. Oxford University Press, Oxford.

Sigursteinsdóttir RJ, Kristjánsson BK (2005) Parallel evolution, not always so parallel: Comparison of small benthic charr, *Salvelinus alpinus*, from Grímsnes and Thingvallavatn, Iceland. Environmental Biology of Fishes 74: 239-244. doi: 10.1007/s10641-005-0499-2

Skúlason S, Parsons KJ, Svanbäck R, Räsänen K, Ferguson MM, Adams CE, Amundsen PA, Bartels P, Bean CW, Boughman JW, Englund G, Guðbrandsson J, Hooker OE, Hudson AG, Kahilainen KK, Knudsen R, Kristjánsson BK, Leblanc CAL, Jónsson Z, Öhlund G, Smith C, Snorrason SS (2019) A way forward with eco evo devo: an extended theory of resource polymorphism with postglacial fishes as model systems. Biological Reviews 94: 1786-1808. doi: 10.1111/brv.12534.

Smith TB (1997) Adaptive significance of the mega-billed form in the polymorphic black-bellied seedcracker *Pyrenestes ostrinus*. Nature. 139: 382-387.

Smith TB, Skúlason S (1996) Evolutionary significance of resource polymorphisms in fishes, amphibians, and birds. Annual Review of Ecology, Evolution, and Systematics. 27: 111-133.

Tremblay S, Magnan P (1991) Interactions between two distantly related species, brook trout (*Salvelinus fontinalis*) and white sucker (*Catostomus commersoni*). Canadian Journal of Fisheries and Aquatic Sciences 48: 857-867.

Venables WN, Ripley BD (2002) Modern applied statistics with S, fourth edition. Springer, New York. ISBN 0-387-95457-0

Wetzel RG, Likens GE (2000). Limnological analyses, third edition. Springer-Verlag, New York.

Zuur AF, Ieno EN, Smith GM (2007). Analysing ecological data. Springer, New York.

Tables

Table 1. Lake characteristics and sampling effort for the 18 lakes sampled. For each lake, the number of brook charr sampled is presented for both the littoral and pelagic zones (L/P). Only creek chub and white sucker were used to calculate mean competitor biomass. Sampling date indicates the first day of sampling; sampling lasted four (2013, 2014) or five (2012) days.

Lake	Sampling dates	Fish community*	Area (ha)	Mean depth (m)	Littoral zone proportion (%)	Brook charr mean biomass (g/trap) (L/P)	Mean competitor biomass (g/trap)	No. of brook charr (L/P)
Baie Cobb (C)	03 + 20-Jul-2012		62	5.88	8	286/896	0	20/20
Maréchal (D)	10 + 17-Jul-2012		103	5.18	10	508/3797	0	19/20
Vertnez (E)	20-May-2013	SEAT	-	-	-	2683/401	138	20/20
Bucheron (G)	03-Jun-2013		10	3.90	18	2275/345	0	19/20
Cutaway (H)	10-Jun-2013	CHEO, SEAT, CACO	40	3.38	15	1885/850	5829	20/20
Visons (J)	24-Jun-2013	CHEO, SEAT	74	7.54	6	939/1533	154	20/20
Jimmy (K)	02-Jul-2013	CHEO, SEAT, CACO, LUCO	-	-	-	910/1030	4718	20/20
Chute noire (N)	22-Jul-2013	CHEO	17	3.80	18	1120/591	0	20/20
Corneille (O)	29-Jul-2013	CHEO, SEAT, CACO	23	4.78	17	316/1241	4891	16/20
Coteau (Q)	20-May-2014	CHEO	31	4.72	16	3137/499	0	20/17

Table 1 (Continued)

Lake	Sampling dates	Fish community*	Area (ha)	Mean depth (m)	Littoral zone proportion (%)	Brook charr mean biomass (g/trap) (L/P)	Mean competitor biomass (g/trap)	No. of brook charr (L/P)
Oudiette (R)	27-May-2014	CHEO, SEAT, CACO	44	8.59	7	1227/336	2180	20/14
Gauthier (S)	03-Jun-2014	CHEO, SEAT, MAMA	37	8.62	9	1327/781	2845	20/20
Shawinigan (T)	10-Jun-2014	CHEO, SEAT, LUCO, MAMA	60	11.70	4	1221/739	1697	20/20
Adam (U)	17-Jun-2014		14	5.24	13	1651/1229	0	18/20
Marshall (W)	01-Jul-2014	SEAT	41	6.39	10	855/1330	3632	16/20
Marmotte (X)	08-Jul-2014	SEAT	14	2.97	27	921/2407	3195	20/19
Plouf (Z)	22-Jul-2014	CHEO, SEAT, CACO, PIPR	60	8.82	5	633/1230	7994	20/20
Simpson (BB)	05-Aug-2014		30	4.24	26	1280/1387	0	15/20

* Fish species codes: SEAT, creek chub (*Semotilus atromaculatus*), CHEO, northern redbelly dace (*Chrosomus eos*), CACO, white sucker (*Catostomus commersonii*), LUCO, common shiner (*Luxilus cornutus*), MAMA, Allegheny pearl dace (Margariscus margarita), COPL, lake chub (*Couesius plumbeus*), CUIN, brook stickleback (*Culaea inconstans*), RHCA, longnose dace (*Rhinichthys cataractae*), PIPR, fathead minnow (*Pimephales promelas*). Brook charr was present in all lakes.

Table 2. Jackknife-based classification for each specific Ecotype × Fish community based on a) morphological traits, b) male body shape, and c) female body shape. The proportion of correct classifications is provided in the last column. Note that a random classification would have been ~16.7% for each group. Global correct classifications were 35.8%, 33.6%, and 32.4% for morphological traits, male body shape, and female body shape, respectively.

Observed	Predicted						Proportion (%)
	Litt. BC	Litt. BC+CC	Litt. BC+CC+WS	Pela. BC	Pela. BC+CC	Pela BC+CC+WS	
A							
Litt. BC	**43**	14	16	35	11	12	32.8
Litt. BC+CC	13	**47**	3	13	16	1	50.5
Litt. BC+CC+WS	15	1	**26**	17	5	50	22.8
Pela. BC	37	11	14	**46**	13	16	33.6
Pela. BC+CC	18	23	6	17	**23**	5	25.0
Pela. BC+CC+WS	11	1	28	12	5	**60**	51.3
B							
Litt. BC	**17**	2	8	16	8	10	27.9
Litt. BC+CC	4	**16**	4	10	19	8	26.2
Litt. BC+CC+WS	6	4	**18**	5	4	10	38.3
Pela. BC	18	6	5	**24**	7	5	36.9
Pela. BC+CC	5	17	5	7	**24**	1	40.7
Pela. BC+CC+WS	9	7	10	4	3	**16**	32.7
C							
Litt. BC	**24**	4	4	23	11	7	32.9
Litt. BC+CC	8	**10**	6	4	23	3	18.5
Litt. BC+CC+WS	7	4	**14**	7	3	10	31.1
Pela. BC	23	8	2	**30**	1	7	42.3
Pela. BC+CC	12	15	0	6	**23**	3	39.0
Pela. BC+CC+WS	9	5	6	11	2	**11**	25.0

Table 3. Pairwise comparisons in magnitude differences (Δd). Upper 95% confidence limit, *Z*-scores, and *p*-values are provided. *Z*-scores and *p*-values are based on 1 000 random permutations. Significant pairwise comparisons ($p < 0.05$) are shown in bold.

Lake comparison	Δd	UCL (95%)	Z	Pr > d
BB:C	0.00106	0.00460	-0.55187	0.64800
BB:D	0.00097	0.00438	-0.56044	0.63300
BB:E	0.00170	0.00444	-0.05027	0.43500
BB:G	0.00228	0.00456	0.33830	0.31400
BB:H	0.00079	0.00459	-0.72692	0.72500
BB:J	0.00018	0.00466	-1.12798	0.92700
BB:K	0.00025	0.00470	-1.11584	0.90500
BB:N	0.00062	0.00445	-0.87491	0.78100
BB:O	0.00290	0.00440	0.75324	0.21300
BB:Q	0.00069	0.00476	-0.79522	0.75500
BB:R	**0.00467**	**0.00466**	**1.96632**	**0.04900**
BB:S	0.00009	0.00467	-1.25809	0.98300
BB:T	0.00059	0.00457	-0.88726	0.81700
BB:U	0.00102	0.00466	-0.53182	0.62900
BB:W	0.00386	0.00463	1.35258	0.10200
BB:X	0.00103	0.00438	-0.54082	0.63300
BB:Z	0.00148	0.00437	-0.20720	0.50300
C:D	0.00009	0.00424	-1.19468	0.95600
C:E	0.00276	0.00435	0.70347	0.22100
C:G	0.00123	0.00454	-0.36635	0.57600
C:H	0.00026	0.00423	-1.13416	0.89500
C:J	0.00087	0.00450	-0.63019	0.67100
C:K	0.00130	0.00439	-0.31553	0.54500
C:N	0.00168	0.00444	-0.07782	0.44000
C:O	0.00396	0.00480	1.42117	0.09900
C:Q	0.00037	0.00448	-1.01439	0.86500
C:R	**0.00573**	**0.00476**	**2.71652**	**0.01800**
C:S	0.00114	0.00440	-0.44740	0.61600
C:T	0.00165	0.00442	-0.06590	0.44400
C:U	0.00208	0.00449	0.25649	0.32400
C:W	**0.00491**	**0.00477**	**2.10276**	**0.04500**
C:X	0.00002	0.00446	-1.26018	0.98100
C:Z	0.00042	0.00452	-0.93492	0.82900
D:E	0.00267	0.00412	0.75599	0.20500

Table 3 (Continued)

Lake comparison	Δ*d*	UCL (95%)	Z	Pr > d
D:G	0.00131	0.00439	-0.30436	0.54600
D:H	0.00017	0.00424	-1.09369	0.93400
D:J	0.00079	0.00453	-0.68302	0.70300
D:K	0.00122	0.00407	-0.33857	0.54300
D:N	0.00159	0.00417	-0.06012	0.43800
D:O	0.00387	0.00433	1.52107	0.07700
D:Q	0.00028	0.00429	-1.04779	0.88000
D:R	**0.00564**	**0.00466**	**2.67686**	**0.01600**
D:S	0.00106	0.00424	-0.40033	0.57200
D:T	0.00156	0.00411	-0.10713	0.45700
D:U	0.00199	0.00425	0.21425	0.34500
D:W	**0.00483**	**0.00427**	**2.17576**	**0.03400**
D:X	0.00006	0.00414	-1.23631	0.97000
D:Z	0.00051	0.00413	-0.86000	0.79500
E:G	0.00399	0.00443	1.55949	0.07700
E:H	0.00250	0.00431	0.57054	0.24200
E:J	0.00189	0.00431	0.14115	0.36500
E:K	0.00146	0.00420	-0.17343	0.50500
E:N	0.00108	0.00444	-0.45772	0.60200
E:O	0.00120	0.00472	-0.42493	0.58300
E:Q	0.00240	0.00443	0.50785	0.25600
E:R	0.00297	0.00466	0.78340	0.19100
E:S	0.00162	0.00462	-0.11048	0.44900
E:T	0.00112	0.00432	-0.48908	0.63700
E:U	0.00068	0.00417	-0.77045	0.75100
E:W	0.00215	0.00476	0.23482	0.32600
E:X	0.00274	0.00437	0.72314	0.19900
E:Z	0.00318	0.00432	1.05828	0.13400
G:H	0.00149	0.00440	-0.17383	0.47600
G:J	0.00210	0.00466	0.21104	0.32800
G:K	0.00253	0.00451	0.59244	0.23900
G:N	0.00290	0.00429	0.87475	0.17500
G:O	**0.00519**	**0.00474**	**2.33525**	**0.03100**
G:Q	0.00159	0.00446	-0.11127	0.47200
G:R	**0.00696**	**0.00460**	**3.54477**	**0.00600**
G:S	0.00237	0.00415	0.52710	0.25900
G:T	0.00287	0.00468	0.79439	0.19900
G:U	0.00331	0.00465	1.04566	0.14300

Table 3 (Continued)

Lake comparison	Δ*d*	UCL (95%)	Z	Pr > d
G:W	**0.00614**	**0.00462**	**2.99782**	**0.01400**
G:X	0.00125	0.00445	-0.37968	0.56300
G:Z	0.00081	0.00435	-0.66035	0.69200
H:J	0.00061	0.00433	-0.83240	0.76600
H:K	0.00104	0.00433	-0.50883	0.63100
H:N	0.00142	0.00442	-0.25575	0.51500
H:O	0.00370	0.00463	1.29818	0.11300
H:Q	0.00010	0.00440	-1.21634	0.95800
H:R	**0.00547**	**0.00465**	**2.52002**	**0.02700**
H:S	0.00088	0.00426	-0.61769	0.67300
H:T	0.00138	0.00443	-0.32012	0.55600
H:U	0.00182	0.00439	0.08740	0.37500
H:W	0.00465	0.00465	2.01642	0.05100
H:X	0.00024	0.00422	-1.10069	0.91000
H:Z	0.00068	0.00444	-0.74684	0.73400
J:K	0.00043	0.00417	-0.98240	0.84500
J:N	0.00080	0.00435	-0.70309	0.71100
J:O	0.00309	0.00454	0.89689	0.16800
J:Q	0.00051	0.00395	-0.91600	0.80800
J:R	**0.00486**	**0.00463**	**2.05249**	**0.04300**
J:S	0.00027	0.00428	-1.10634	0.91000
J:T	0.00077	0.00435	-0.71901	0.72700
J:U	0.00121	0.00409	-0.32457	0.53700
J:W	0.00404	0.00453	1.54141	0.08400
J:X	0.00085	0.00430	-0.62664	0.66900
J:Z	0.00129	0.00422	-0.30936	0.53800
K:N	0.00037	0.00427	-1.02068	0.85700
K:O	0.00266	0.00451	0.62068	0.23200
K:Q	0.00094	0.00422	-0.59005	0.66000
K:R	0.00443	0.00451	1.87301	0.05400
K:S	0.00016	0.00454	-1.16552	0.93000
K:T	0.00034	0.00446	-1.02985	0.87300
K:U	0.00078	0.00416	-0.75195	0.73100
K:W	0.00361	0.00470	1.21655	0.12000
K:X	0.00128	0.00426	-0.25469	0.50700
K:Z	0.00172	0.00408	0.02950	0.42000
N:O	0.00228	0.00457	0.29916	0.31800
N:Q	0.00131	0.00441	-0.29346	0.53300

Table 3 (Continued)

Lake comparison	Δd	UCL (95%)	Z	Pr > d
N:R	0.00405	0.00448	1.54342	0.08200
N:S	0.00053	0.00441	-0.88556	0.77600
N:T	0.00003	0.00428	-1.30686	0.98700
N:U	0.00040	0.00436	-0.98757	0.86000
N:W	0.00324	0.00448	1.00950	0.14600
N:X	0.00165	0.00429	-0.07049	0.43500
N:Z	0.00210	0.00433	0.29206	0.32700
O:Q	0.00359	0.00453	1.26502	0.11500
O:R	0.00177	0.00447	-0.02490	0.41900
O:S	0.00282	0.00463	0.69411	0.21000
O:T	0.00231	0.00460	0.31100	0.31600
O:U	0.00188	0.00432	0.02671	0.42900
O:W	0.00095	0.00487	-0.59281	0.66600
O:X	0.00394	0.00443	1.47855	0.08300
O:Z	0.00438	0.00459	1.75140	0.06300
Q:R	**0.00536**	**0.00459**	**2.46669**	**0.02800**
Q:S	0.00078	0.00444	-0.70325	0.71100
Q:T	0.00128	0.00444	-0.34607	0.54700
Q:U	0.00171	0.00441	-0.05473	0.43700
Q:W	**0.00455**	**0.00454**	**1.91015**	**0.04900**
Q:X	0.00034	0.00436	-1.00457	0.86500
Q:Z	0.00079	0.00452	-0.64832	0.69300
R:S	0.00459	0.00470	1.81459	0.05700
R:T	0.00408	0.00482	1.49745	0.08800
R:U	0.00365	0.00451	1.24349	0.10900
R:W	0.00082	0.00484	-0.68301	0.70300

Table 3 (Continued)

Lake comparison	Δd	UCL (95%)	Z	Pr > d
R:X	**0.00571**	**0.00452**	**2.78306**	**0.01500**
R:Z	**0.00615**	**0.00478**	**2.91932**	**0.01300**
S:T	0.00050	0.00421	-0.94096	0.81300
S:U	0.00094	0.00427	-0.61249	0.67300
S:W	0.00377	0.00481	1.34016	0.10000
S:X	0.00112	0.00423	-0.43190	0.58700
S:Z	0.00156	0.00436	-0.15328	0.48400
T:U	0.00043	0.00459	-0.95604	0.83200
T:W	0.00327	0.00458	1.02731	0.14700
T:X	0.00162	0.00447	-0.08872	0.46000
T:Z	0.00207	0.00443	0.25237	0.32900
U:W	0.00283	0.00461	0.69008	0.20900
U:X	0.00206	0.00414	0.25028	0.33300
U:Z	0.00250	0.00422	0.61434	0.21600
W:X	**0.00489**	**0.00451**	**2.08119**	**0.03900**
W:Z	**0.00533**	**0.00451**	**2.46491**	**0.02600**
X:Z	0.00044	0.00424	-0.93156	0.83000

Table 4. Pairwise comparisons in direction differences. Correlations between trajectories (r), observed values (angle in degrees), upper 95% confidence limit, Z-scores, and p-values are provided. Z-scores and p-values are based on 1 000 random permutations. Significant pairwise comparisons ($p < 0.05$) are shown in bold.

Lake comparison	r	Angle (°)	UCL (95%)	Z	Pr > angle
BB:C	-0.18881	100.88318	113.68129	1.06413	0.15700
BB:D	-0.37132	111.79711	112.57252	1.63749	0.05500
BB:E	**-0.71410**	**135.56975**	**109.79463**	**3.14494**	**0.00100**
BB:G	**-0.50451**	**120.29850**	**109.90697**	**2.31217**	**0.01500**
BB:H	-0.11038	96.33750	111.50394	0.82310	0.22400
BB:J	-0.23766	103.74874	109.74494	1.31451	0.09800
BB:K	0.47584	61.58591	112.74024	-1.12971	0.88100
BB:N	0.30730	72.10358	111.25979	-0.48992	0.68300
BB:O	-0.19817	101.43008	112.65033	1.04947	0.14400
BB:Q	0.08152	85.32386	113.88756	0.17689	0.41900
BB:R	0.10364	84.05129	114.23860	0.10808	0.44000
BB:S	-0.29087	106.91033	110.60074	1.43501	0.08100
BB:T	0.44794	63.38823	109.85648	-1.01818	0.83300
BB:U	-0.38955	112.92648	112.92958	1.71405	0.05100
BB:W	**-0.51984**	**121.32126**	**112.63572**	**2.18466**	**0.01800**
BB:X	0.39834	66.52563	112.94817	-0.81039	0.77900
BB:Z	-0.17055	99.81960	110.96814	1.00412	0.17100
C:D	-0.08530	94.89334	112.65091	0.73930	0.24200
C:E	0.10017	84.25090	110.89189	0.19363	0.41200
C:G	0.33196	70.61226	110.92951	-0.55740	0.70600
C:H	0.18325	79.44070	110.14430	-0.08668	0.53800
C:J	0.28122	73.66720	111.22959	-0.43633	0.65200
C:K	-0.15721	99.04484	112.98038	0.91627	0.18400
C:N	0.11904	83.16359	111.91507	0.05912	0.48800
C:O	0.50466	59.69093	113.83984	-1.17035	0.87500
C:Q	0.04955	87.16008	112.01218	0.33588	0.35100
C:R	0.14462	81.68493	112.80607	0.01855	0.49600
C:S	0.53844	57.42231	111.42030	-1.24582	0.89400
C:T	**-0.37434**	**111.98352**	**109.61964**	**1.79237**	**0.04300**

Table 4 (Continued)

Lake comparison	r	Angle (°)	UCL (95%)	Z	Pr > angle
C:U	0.09013	84.82899	112.09232	0.22293	0.39600
C:W	0.35836	69.00054	110.45354	-0.71825	0.75800
C:X	-0.01014	90.58093	109.95097	0.58953	0.28600
C:Z	0.27757	73.88485	108.67926	-0.37130	0.62400
D:E	-0.02348	91.34546	110.80406	0.60651	0.27100
D:G	0.22053	77.25989	111.74618	-0.22363	0.56800
D:H	-0.18694	100.77405	112.52745	1.05470	0.14500
D:J	0.04795	87.25171	112.27465	0.30135	0.37800
D:K	-0.30385	107.68926	112.28136	1.43902	0.07900
D:N	-0.21512	102.42271	108.71441	1.21538	0.11400
D:O	-0.29961	107.43388	112.09055	1.45689	0.08100
D:Q	-0.35356	110.70550	112.32065	1.66279	0.06200
D:R	-0.10853	96.23053	112.28703	0.82314	0.20700
D:S	-0.18357	100.57792	111.29111	1.13159	0.13500
D:T	-0.30554	107.79051	112.36390	1.44231	0.08900
D:U	0.33187	70.61794	111.43096	-0.60310	0.71000
D:W	0.12840	82.62292	113.12630	0.04102	0.47200
D:X	0.17013	80.20478	112.57546	0.03458	0.47200
D:Z	-0.18938	100.91635	111.99951	1.09587	0.14200
E:G	0.45428	62.98155	109.65502	-1.00729	0.83700
E:H	0.13974	81.96703	109.30565	0.11640	0.44600
E:J	0.25575	75.18206	112.07332	-0.33503	0.62900
E:K	-0.42266	115.00238	110.64783	1.93152	0.02900
E:N	-0.23394	103.52889	113.67488	1.21307	0.13000
E:O	0.21751	77.43688	112.04018	-0.18483	0.55400
E:Q	-0.05713	93.27527	111.40044	0.65932	0.25000
E:R	-0.12264	97.04455	110.47321	0.89397	0.20100
E:S	0.18790	79.16962	109.81492	-0.03621	0.52200
E:T	-0.29419	107.10919	110.74319	1.41416	0.08600
E:U	0.18485	79.34778	112.20377	-0.11832	0.52600
E:W	0.53790	57.45916	110.30097	-1.30745	0.89900
E:X	**-0.54144**	**122.78149**	**110.66938**	**2.38078**	**0.00700**
E:Z	0.28867	73.22166	110.14989	-0.42199	0.64900
G:H	0.21856	77.37528	113.26163	-0.24892	0.58200
G:J	0.14941	81.40701	110.98765	0.01407	0.46700
G:K	-0.15805	99.09375	111.56167	0.97967	0.18100

Table 4 (Continued)

Lake comparison	r	Angle (°)	UCL (95%)	Z	Pr > angle
G:N	-0.06912	93.96339	111.53142	0.73186	0.22700
G:O	0.31633	71.55869	112.51921	-0.55216	0.69100
G:Q	-0.14276	98.20768	112.05894	0.94529	0.18000
G:R	-0.17112	99.85279	115.38391	0.94653	0.18500
G:S	0.39889	66.49097	111.49099	-0.78829	0.77700
G:T	**-0.44974**	**116.72727**	**112.07437**	**1.95002**	**0.03700**
G:U	0.15508	81.07849	111.89513	-0.02429	0.50000
G:W	0.54253	57.14382	111.99497	-1.38330	0.93000
G:X	-0.07376	94.22997	111.47300	0.77180	0.22400
G:Z	0.12362	82.89906	110.08700	0.11752	0.46300
H:J	0.21859	77.37391	113.21182	-0.20420	0.55100
H:K	0.24730	75.68215	111.15283	-0.37416	0.64100
H:N	0.30832	72.04174	111.51388	-0.48574	0.67000
H:O	0.27906	73.79587	113.51562	-0.44911	0.65100
H:Q	0.05253	86.98898	112.70081	0.24371	0.40600
H:R	-0.13616	97.82565	112.04849	0.91167	0.19000
H:S	-0.16227	99.33854	111.17661	1.06138	0.15700
H:T	0.35191	69.39560	113.68068	-0.65332	0.72900
H:U	0.66754	48.12263	112.56988	-1.77593	0.96500
H:W	0.47022	61.95161	111.74263	-1.05110	0.85600
H:X	-0.14705	98.45601	109.59245	1.03052	0.16500
H:Z	0.41519	65.46843	112.27842	-0.82920	0.79400
J:K	-0.00252	90.14422	112.88479	0.44848	0.33200
J:N	0.22186	77.18179	113.33078	-0.28431	0.60500
J:O	0.64174	50.07817	110.14053	-1.64630	0.95500
J:Q	0.01334	89.23585	112.09710	0.41881	0.34500
J:R	0.51767	58.82403	113.37912	-1.25931	0.89100
J:S	0.34148	70.03279	109.81108	-0.61336	0.72000
J:T	-0.27089	105.71724	111.49766	1.34825	0.10300
J:U	0.14564	81.62549	112.60834	0.02023	0.48700
J:W	0.39465	66.75590	111.25158	-0.83035	0.79200
J:X	-0.19411	101.19247	109.61911	1.18798	0.12500
J:Z	0.68436	46.81483	111.22541	-1.87960	0.98000
K:N	0.19174	78.94537	111.53235	-0.16653	0.55600
K:O	-0.01135	90.65042	113.42935	0.45543	0.32000
K:Q	0.48701	60.85573	112.81688	-1.12737	0.86400
K:R	0.39494	66.73797	113.05913	-0.83219	0.79200

Table 4 (Continued)

Lake comparison	r	Angle (°)	UCL (95%)	Z	Pr > angle
K:S	-0.03470	91.98872	112.13660	0.59533	0.27500
K:T	0.11112	83.62037	111.63912	0.09882	0.47200
K:U	-0.17726	100.20996	112.54965	1.05645	0.14000
K:W	**-0.41163**	**114.30747**	**112.26386**	**1.77327**	**0.04200**
K:X	-0.15976	99.19295	112.18788	0.94915	0.16900
K:Z	-0.06439	93.69171	111.93144	0.69067	0.25200
N:O	0.34733	69.67564	111.52433	-0.63561	0.72700
N:Q	0.26774	74.47010	112.05000	-0.34623	0.61300
N:R	0.01430	89.18087	113.08229	0.44925	0.31300
N:S	-0.19310	101.13371	109.57861	1.15173	0.13200
N:T	0.45273	63.08102	108.92718	-1.01354	0.82600
N:U	0.10358	84.05438	110.77246	0.17550	0.42200
N:W	-0.08822	95.06145	110.80996	0.78545	0.22100
N:X	0.18488	79.34578	110.75482	-0.10513	0.53300
N:Z	-0.01237	90.70871	110.52455	0.55319	0.29900
O:Q	0.00290	89.83358	111.25101	0.47091	0.31200
O:R	0.19191	78.93598	111.58401	-0.13507	0.55400
O:S	0.52252	58.49830	112.29078	-1.23337	0.89000
O:T	-0.18440	100.62603	112.42337	1.03851	0.15100
O:U	-0.01268	90.72651	111.08590	0.51456	0.29600
O:W	0.39893	66.48859	110.91495	-0.85785	0.79100
O:X	-0.27528	105.97891	109.81580	1.40336	0.08500
O:Z	0.61386	52.13111	111.44367	-1.57852	0.93300
Q:R	0.57123	55.16391	111.41136	-1.43980	0.92900
Q:S	0.28269	73.57902	111.29322	-0.37889	0.62100
Q:T	-0.00801	90.45904	112.18691	0.51144	0.32000
Q:U	-0.29211	106.98434	111.89112	1.37823	0.09400
Q:W	**-0.51508**	**121.00298**	**112.84693**	**2.16115**	**0.01600**
Q:X	-0.20982	102.11197	110.60366	1.18450	0.13200
Q:Z	-0.26612	105.43328	111.89933	1.35269	0.09100
R:S	0.46326	62.40235	110.67197	-1.05511	0.85400
R:T	-0.41121	114.28097	110.62876	1.85954	0.03500
R:U	**-0.38210**	**112.46366**	**111.70642**	**1.71563**	**0.04400**
R:W	-0.28622	106.63163	112.37513	1.34050	0.10400
R:X	-0.17761	100.23086	112.02692	1.01830	0.16800

Table 4 (Continued)

Lake comparison	r	Angle (°)	UCL (95%)	Z	Pr > angle
R:Z	0.00199	89.88580	112.93733	0.43659	0.33600
S:T	**-0.65879**	**131.20788**	**109.35039**	**2.83368**	**0.00300**
S:U	**-0.33705**	**109.69747**	**109.35734**	**1.65768**	**0.04700**
S:W	0.14637	81.58313	110.84518	-0.00066	0.49800
S:X	-0.14962	98.60463	109.69673	1.04793	0.15800
S:Z	0.21799	77.40875	111.36395	-0.12453	0.54500
T:U	0.23103	76.64212	112.49355	-0.23503	0.57300
T:W	-0.17231	99.92241	112.14339	1.01716	0.17000
T:X	0.21931	77.33169	110.37965	-0.11575	0.54000
T:Z	-0.04015	92.30112	110.91207	0.68487	0.24900
U:W	0.61384	52.13206	111.72534	-1.63060	0.95200
U:X	-0.08322	94.77375	111.36136	0.82351	0.21800
U:Z	0.25396	75.28810	110.41693	-0.30255	0.60700
W:X	-0.07994	94.58499	111.43027	0.76549	0.23100
W:Z	0.48525	60.97098	109.92989	-1.12210	0.86300
X:Z	-0.23579	103.63821	110.89095	1.33433	0.09200

Table 5. Results of the MANOVA based on body shape. Italicized values indicate where F-values were calculated with respect to random effects (fish community:year) rather than residuals. Z and *p* are based on 1,000 random permutations using the residual randomization in permutation procedure (RRPP).

Module	Df	Sum of squares	MS	R2	F	Z	*p*-value
Csize	1	0.021	0.0211	0.055	54.93	8.30	0.001
Sex	1	0.011	0.011	0.023	28.89	7.47	0.001
Ecotype	1	0.001	0.001	0.003	2.99	2.67	0.004
Fish community	2	0.012	0.006	0.032	*1.13*	*0.37*	*0.361*
Fish community:Lake	15	0.081	0.005	0.213	14.17	17.36	0.001
Residuals	667	0.253	0.001	0.668			
Total	687	0.378					

Figures captions

Figure 1. Schematic drawing showing the 15 morphological traits (dashed blue lines) measured using landmark (red dots) and semi-landmark (blue dots) coordinates. AB, anal fin base; AH, anal fin height; BH, body height; DB, dorsal fin base; DD, dorsal fin; DH, dorsal fin height; HH, head height; HL, head length; PC, pectoral fin; PD, peduncle; PH, pelvic fin height; PS, peduncle height; PL, pelvic fin; SC, snout to posterior cranium; SE, snout to eye; SJ, upper jaw. Red dots represent landmarks used in body shape analysis; blue dots represent semi-landmarks used in morphologic trait analysis only, and green dots represent curve points used to "unbend" specimens prior to shape analysis. In addition, mouth width (MW) and body width (BW) were two morphological traits measured in the field.

Figure 2. Effect size and proportion of phenotypic variation (R2) explained by the ecotype term in the 17 morphological traits of the study lakes (circles). The mean effect size and semi-partial R2 based on mixed models are presented (diamonds). Red symbols represent significant effects of the ecotype term. Positive and negative effect size represents greater trait values for the pelagic and littoral ecotypes, respectively.

Figure 3. Morphological discriminant scores (a) and canonical correlation coefficients (b) on the first two axes of the linear discriminant function analysis (LDA) based on 17 morphological traits of brook charr. Small symbols refer to discriminant scores of individuals and large symbols indicate the centroid of each specific Ecotype × Fish community category. Blue symbols/lines: BC lakes; yellow symbols/lines: BC+CC lakes; brown symbols/lines: BC+CC+WS lakes; circles: littoral ecotype; triangles: pelagic ecotype. Ellipses include 80% of the individuals in each category. Solid lines: littoral ecotype; dashed lines: pelagic ecotype. See Figure 1 for definitions of the morphological traits.

Figure 4. Morphological discriminant scores of (a) male and (b) female brook charr based on 15 body shape landmarks. The first two axes of the linear discriminant function analysis (LDA) are presented. Small symbols refer to discriminant scores of individuals and large symbols indicate the centroid of each specific Ecotype × Fish community category. Blue symbols/lines: BC lakes; yellow symbols/lines: BC+CC lakes; brown symbols/lines: BC+CC+WS lakes; circles: littoral ecotype; triangles: pelagic ecotype. Ellipses include 80% of the individuals in each category. Solid lines: littoral ecotype; dashed lines: pelagic ecotype. The body shape of individuals at the extreme of each axis are presented for axes interpretation. Morphological differences between extreme individuals (black circle) and overall mean body shape (grey points) were magnified three times.

Figure 5. Phenotypic change trajectories along a morphospace represented by the first two PC axes (a) and shape change from the consensus at the extremities of the first two PC axes (b). In (a), grey letters refer to individual fish identified by the name of the lake (see Table 1 for lake identification). Arrows indicate shape change from littoral ecotype to pelagic ecotype for each lake. Colours refer to fish communities. In (b), vector displacements between corresponding landmarks in the consensus (grey symbols) and target specimens (black symbols) are shown to help the interpretation of the phenotypic trajectory analysis.

Figures

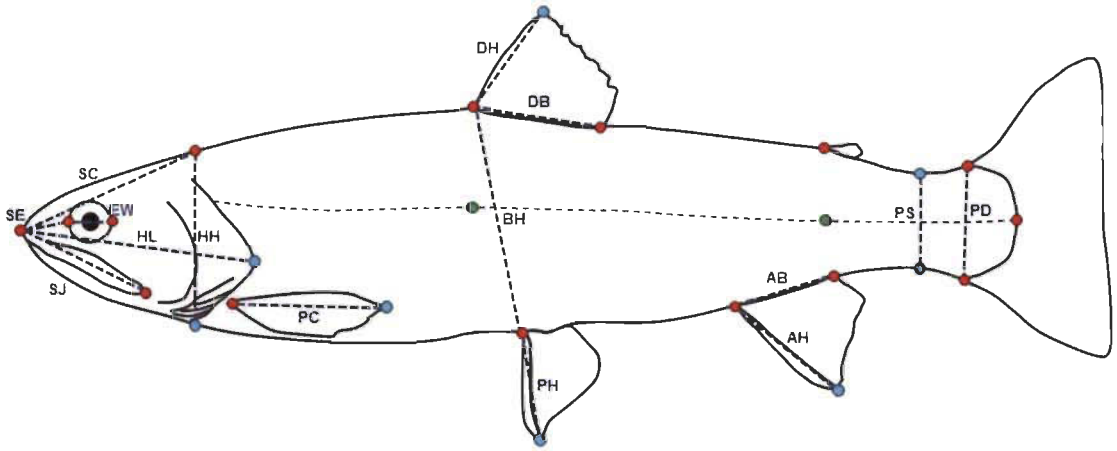

Figure 1.

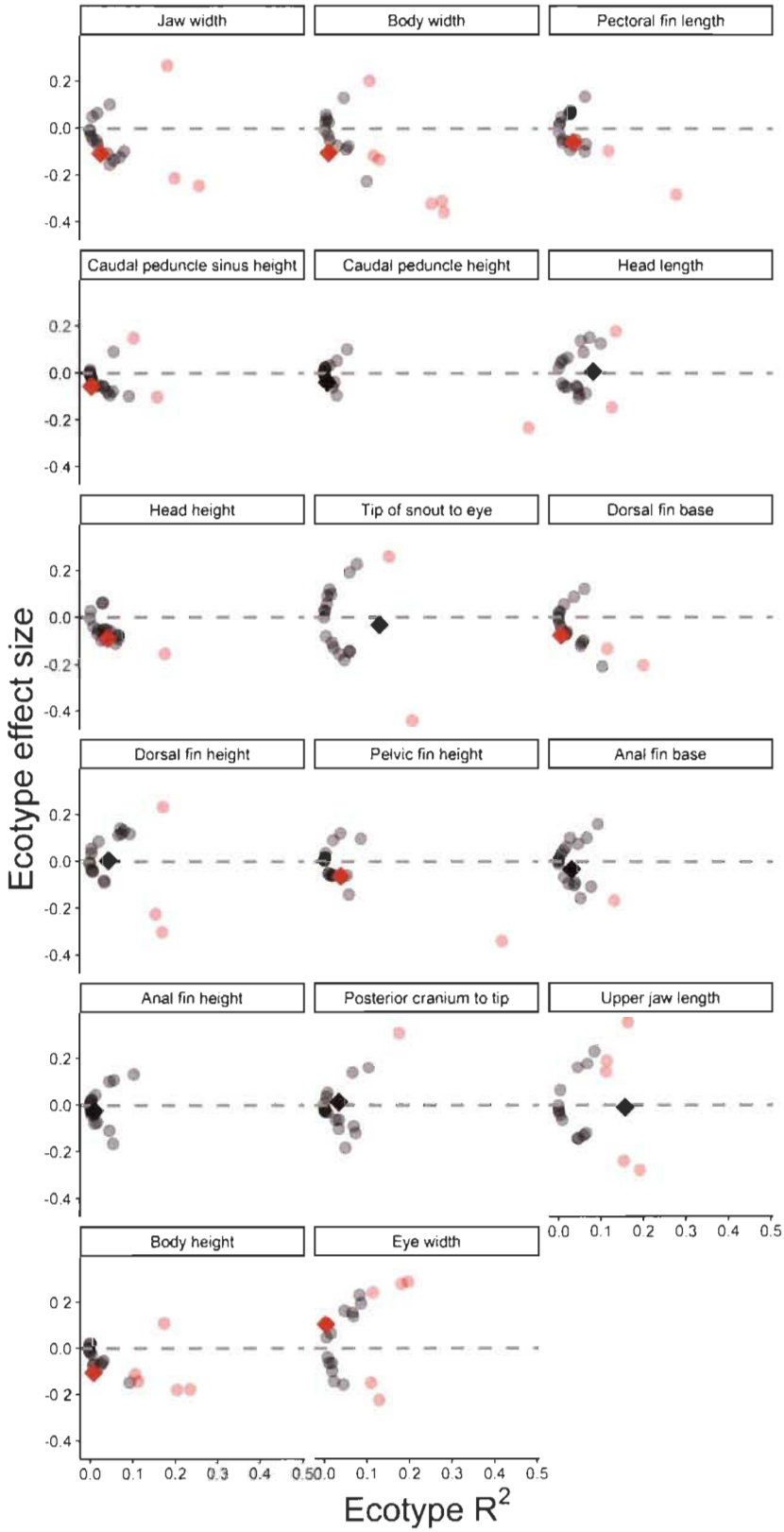
Jaw width
Body width
Pectoral fin length
Caudal peduncle sinus height
Caudal peduncle height
Head length
Head height
Tip of snout to eye
Dorsal fin base
Dorsal fin height
Pelvic fin height
Anal fin base
Anal fin height
Posterior cranium to tip
Upper jaw length
Body height
Eye width
Ecotype effect size
Ecotype R^2

Figure 2.

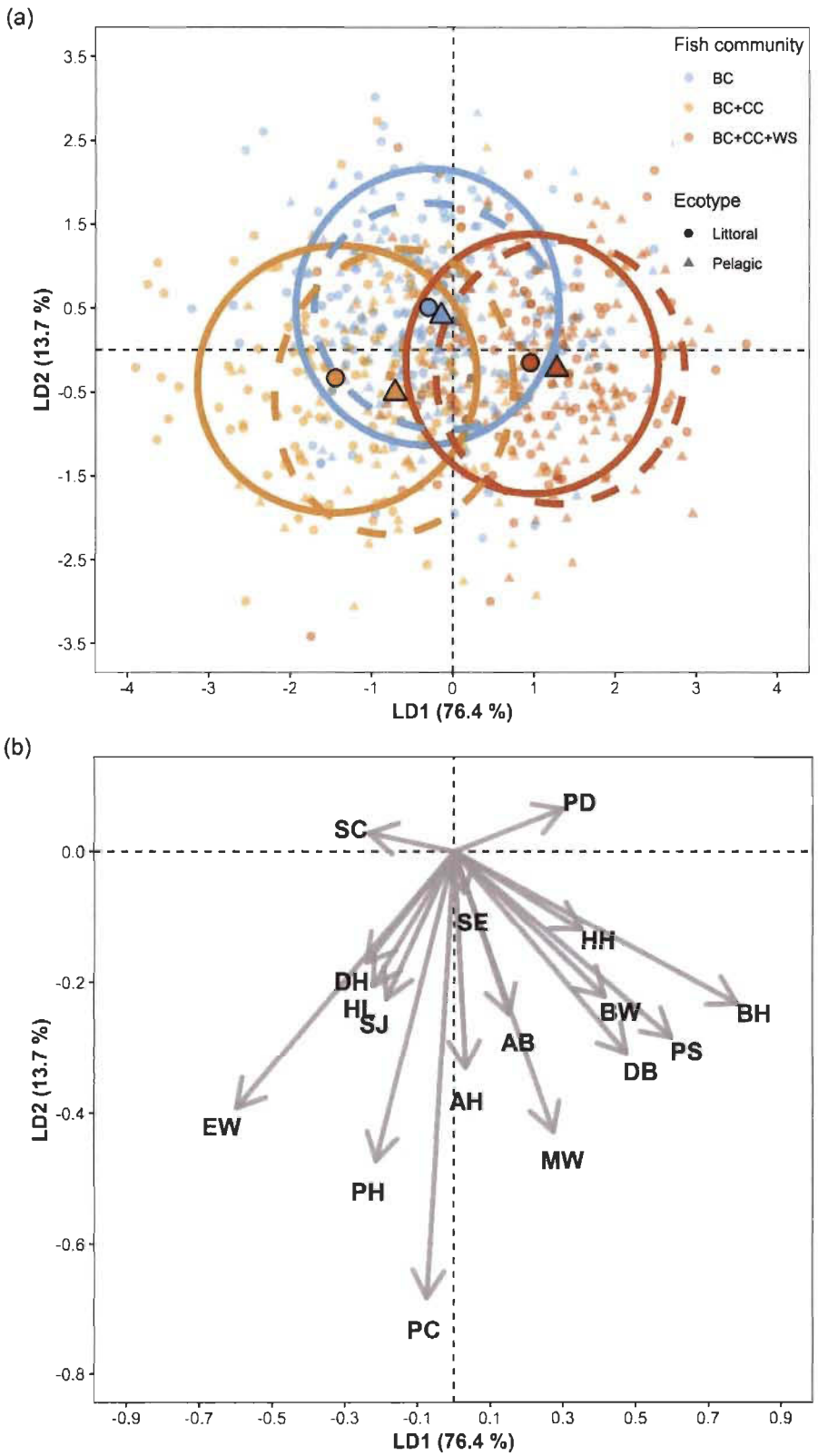
(a)
Fish community
BC
BC+CC
BC+CC+WS
Ecotype
Littoral
Pelagic
LD2 (13.7 %)
LD1 (76.4 %)
(b)
PD
SC
SE
HH
DH
HL
SJ
BW
BH
AB
PS
DB
AH
EW
MW
PH
PC
LD2 (13.7 %)
LD1 (76.4 %)

Figure 3.

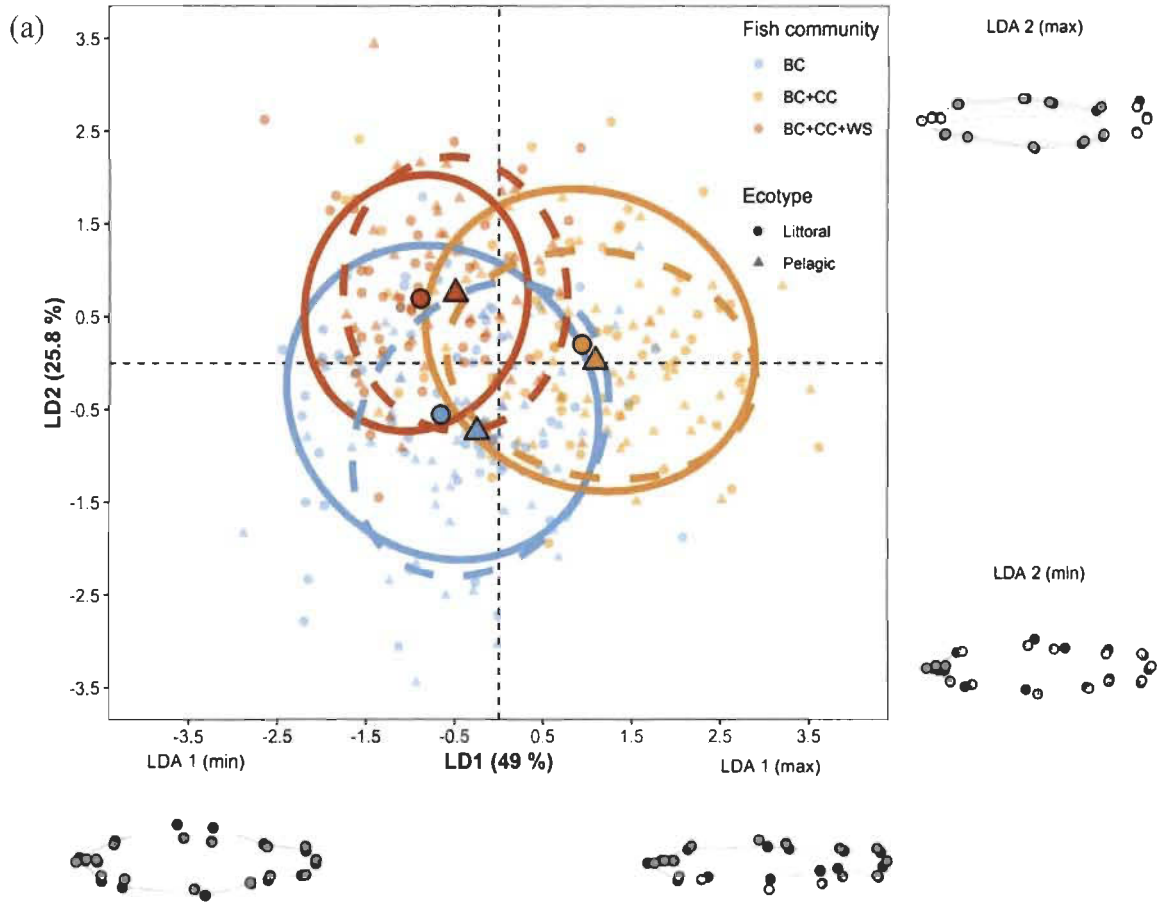
(a)
Fish community
BC
BC+CC
BC+CC+WS
Ecotype
Littoral
Pelagic
LDA 2 (max)
LDA 2 (min)
LD2 (25.8 %)
LD1 (49 %)
LDA 1 (min)
LDA 1 (max)
3.5
2.5
1.5
0.5
-0.5
-1.5
-2.5
-3.5

Figure 4.

(b)

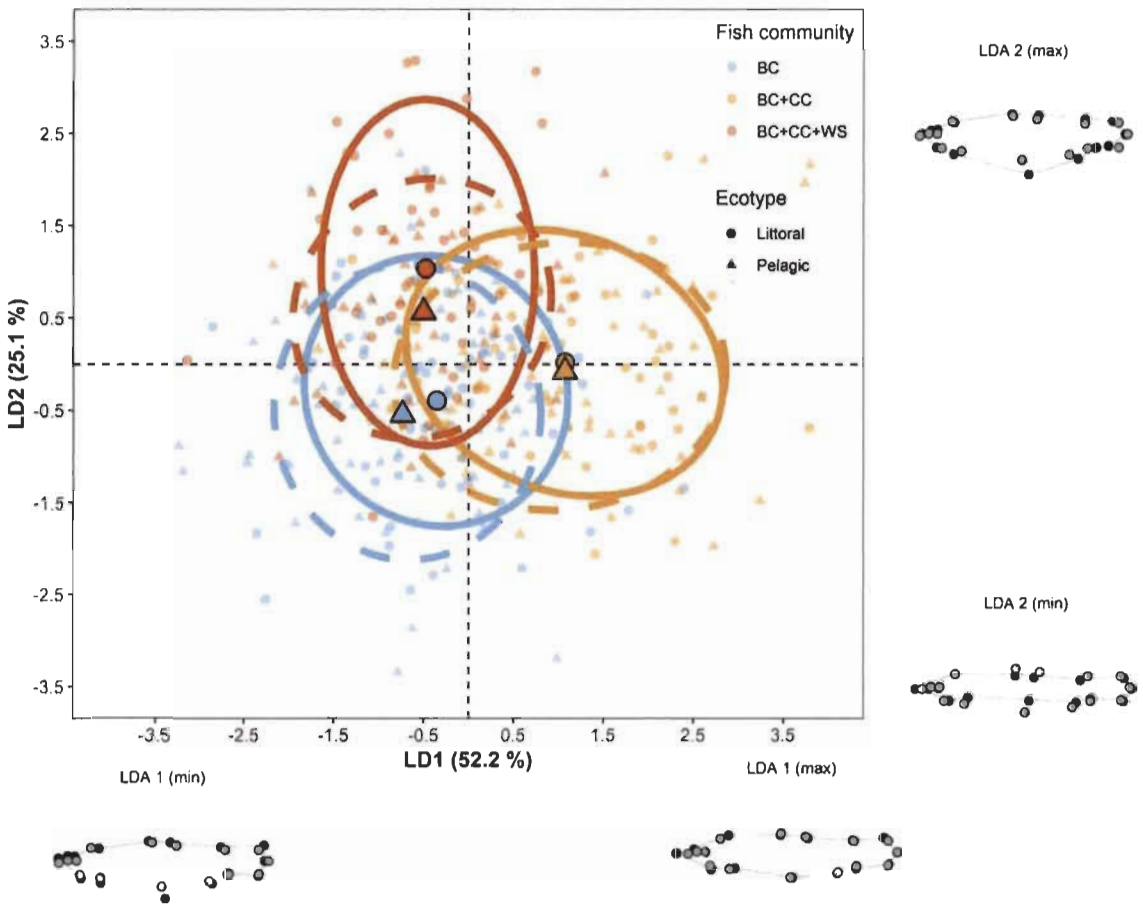

Figure 4 (Continued)

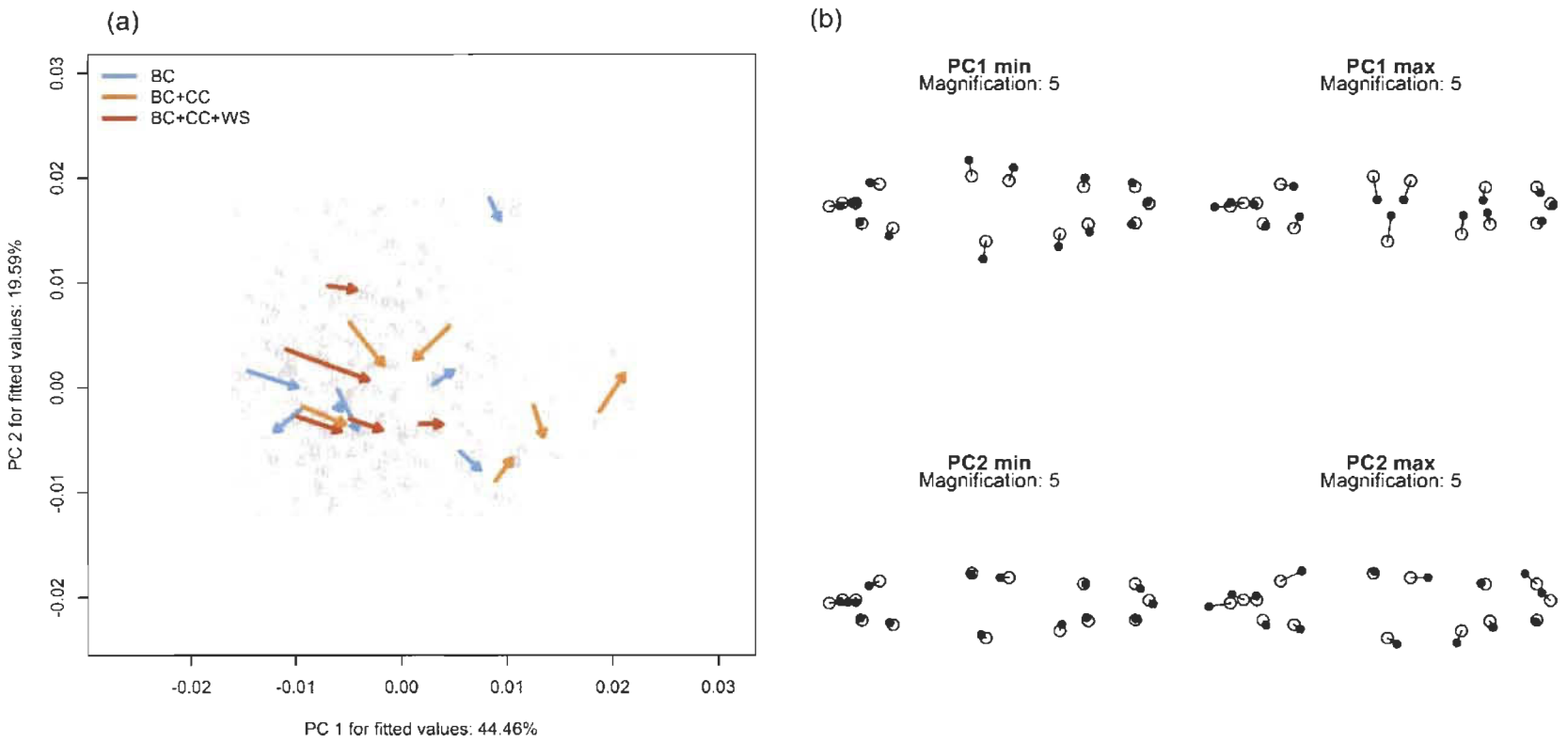
(a)
BC
BC+CC
BC+CC+WS
PC 2 for fitted values: 19.59%
PC 1 for fitted values: 44.46%
-0.02
-0.01
0.00
0.01
0.02
0.03
(b)
PC1 min
Magnification: 5
PC1 max
Magnification: 5
PC2 min
Magnification: 5
PC2 max
Magnification: 5

Figure 5.

CHAPITRE V

DISCUSSION GÉNÉRALE

5.1 Synthèse et contributions scientifiques de la thèse

Cette thèse a examiné l'impact de la compétition interspécifique et de plusieurs facteurs environnementaux sur l'expression du polymorphisme associé aux ressources, sur l'utilisation des ressources ainsi que sur la sélection entre les habitats littoral et pélagique d'une trentaine de populations lacustres d'omble de fontaine indigènes. Nous avons observé que la compétition interspécifique par le meunier noir réduit drastiquement l'abondance des ombles de fontaine, et ce, dans les deux habitats (c.-à-d., littoral et pélagique). Toutefois, nous avons constaté que la présence de meunier noir ne semble pas affecter la relation de densité-dépendance dans la sélection de l'habitat de l'omble de fontaine (Chapitre II). Cette relation de densité-dépendance entre les abondances d'ombles de fontaine en zone littorale et pélagique suggère que la compétition intraspécifique est un facteur important affectant l'utilisation d'habitats dans les populations d'omble de fontaine lacustres (Chapitre II). Nos travaux ont également démontré comment la température de l'eau peut altérer fortement la distribution spatiale des ombles de fontaine entre les habitats en agissant comme une barrière thermique limitant l'accessibilité aux ressources littorales (Chapitre II). Ces travaux ont également permis de confirmer qu'il est commun de retrouver, chez l'omble de fontaine, une spécialisation dans l'utilisation des ressources entre les écotypes littoral et pélagique dans les lacs du Bouclier laurentien et que cette spécialisation est stable dans le temps (Chapitre III). Cependant, contrairement à ce que suggère l'hypothèse de l'opportunité écologique, nous n'avons pas observé une spécialisation alimentaire plus marquée dans les lacs allopatriques comparativement aux populations où l'omble de fontaine subit une pression de compétition interspécifique pour les ressources littorales (c.-à-d., les communautés sympatriques avec mulets à cornes et/ou meuniers noirs). En effet,

en présence de compétiteurs en zone littorale, les deux écotypes semblent plutôt incorporer davantage de ressources pélagiques à leur diète tout en maintenant l'amplitude de leur spécialisation alimentaire (Chapitre III). De plus, en ce qui a trait aux différences morphologiques entre les écotypes, nous avons observé de faibles niveaux d'évolution parallèle entre les populations (Chapitre IV). Dans l'ensemble, l'écotype littoral s'est avéré légèrement plus trapu avec des nageoires plus longues et de plus petits yeux (Chapitre IV). De plus, la compétition interspécifique en zone littorale ne semble pas engendrer de modifications dans l'amplitude des différences morphologiques entre les écotypes littoral et pélagique dans ce système (Chapitre IV). Enfin, les différences morphologiques se sont également avérées plus importantes entre les individus de lacs différents qu'entre les écotypes d'un même lac suggérant un effet important des conditions environnementales locales de chaque lac.

Le chapitre II de cette thèse s'est d'abord attardé à évaluer l'influence de la compétition interspécifique ainsi que de plusieurs facteurs environnementaux (c.-à-d., la température de l'épilimnion, la proportion de la zone littorale et la saisonnalité) sur l'utilisation des habitats littoral et pélagique par l'omble de fontaine. Pour ce faire, nous avons évalué l'effet de ces différents facteurs environnementaux sur l'abondance des ombles de fontaine dans les habitats littoral et pélagique indépendamment. Puis, nous avons développé un cadre théorique basé sur les isodars (Morris 1988) nous permettant, entre autres, de déterminer le ou les habitats préférentiels, si cette préférence est dépendante de la densité (c.-à-d., si la compétition intraspécifique affecte la sélection d'habitats) et comment la compétition interspécifique ainsi que certains facteurs environnementaux affectent les patrons spatiaux d'abondance de l'omble de fontaine lacustre. Nos résultats montrent un effet clair de la compétition interspécifique par le meunier noir sur l'abondance des ombles de fontaine autant en zone littorale que pélagique, entraînant une diminution des abondances de près de 50% comparativement aux populations allopatriques. Toutefois, aucun effet sur l'abondance des ombles de fontaine n'a été détecté en réponse à la présence de mulet à cornes. Une contribution importante de ce chapitre aura été d'avoir mis en lumière un effet de densité-dépendance dans la sélection d'habitats, un phénomène souvent cité comme favorisant l'expression du

polymorphisme associé aux ressources via la plasticité phénotypique et la sélection divergente (Smith and Skúlason 1996; Svanbäck and Bolnick 2007; Martin and Pfennig 2010; Araújo et al. 2011; Skúlason et al. 2019). Nos résultats ont montré que les ombles de fontaine préfèrent l'habitat littoral lorsque la température de l'épilimnion est optimale (~16°C). Ces analyses montrent également que l'augmentation de la température de l'épilimnion affecte drastiquement cette relation de densité-dépendance et que les ombles évitent l'habitat littoral lorsque la température de l'épilimnion dépasse 22.2°C. Cet effet marqué de la température de l'épilimnion sur la sélection d'habitats des ombles de fontaine rappelle l'importance d'inclure la qualité thermique des habitats dans l'étude de la sélection d'habitats chez les ectothermes (Halliday et Blouin-Demer 2017). Considérant que les changements climatiques devraient engendrer une augmentation de la température des eaux de surface de plusieurs degrés au cours du siècle présent (O'Reilly et al. 2015; Woolway and Merchant 2019), les changements climatiques pourraient donc limiter l'effet de diversification de la compétition intraspécifique et restreindre l'expression du polymorphisme associé aux ressources.

L'objectif du chapitre III a été de tester l'hypothèse de l'opportunité écologique dans ce système en s'attardant particulièrement à l'utilisation des ressources littorales et pélagiques par l'omble de fontaine, et ce, en réponse à différents niveaux de compétition interspécifique. Une opportunité écologique se présente lorsqu'une ressource devient disponible pour une population suite à la colonisation d'un nouvel habitat, au relâchement de la compétition interspécifique ou de la prédation et que cette niche vacante est colonisée par ladite population (Schluter 2000, Yoder et al. 2010). Il est suggéré que la plasticité phénotypique et la compétition intraspécifique sont les principaux facteurs induisant la diversification de l'utilisation des ressources et que ces phénomènes seraient impliqués dans l'expression du polymorphisme associé aux ressources (Martin et Pfennig 2010, Nosil et Reimchen 2005). Selon l'hypothèse de l'opportunité écologique, nous devrions observer une plus grande spécialisation entre les écotypes dans les lacs où la compétition interspécifique est faible/absente (Yoder et al. 2010). Pour vérifier cette hypothèse, cinq indicateurs de l'utilisation des ressources présentant différents temps d'intégration (c.-à-d., contenus stomacaux, signature isotopique du carbone dans le foie, concentration

en caroténoïdes dans les muscles, longueur des caeca pyloriques et longueur des branchicténies) ont été analysés. Nos résultats montrent une spécialisation alimentaire claire des deux écotypes pour tous les indicateurs, excepté pour la longueur des branchicténies, suggérant que ces spécialisations sont stables dans le temps. De plus, tous les indicateurs, sauf la signature isotopique du foie, indiquent que la compétition interspécifique pousse les ombles à incorporer davantage de ressources pélagiques dans leur alimentation. Cependant, contrairement à notre hypothèse de départ, la pression de compétition interspécifique ne semble pas affecter le degré de spécialisation entre les deux écotypes. Nos résultats suggèrent plutôt que les interactions entre des espèces phylogénétiquement distantes ne permettent pas une exclusion complète de l'omble de fontaine de la zone littorale, expliquant ainsi l'absence de relâchement écologique en allopatrie (c.-à-d., l'absence de différences plus grandes dans l'utilisation des ressources entre les deux écotypes dans les lacs allopatriques).

L'objectif du chapitre IV a été de vérifier quels sont les traits morphologiques impliqués chez l'omble de fontaine dans la spécialisation aux habitats littoral et pélagique et de vérifier si différentes populations présentent une évolution parallèle de ces deux écotypes. L'évolution parallèle d'écotypes a lieu lorsque les écotypes de populations distinctes présentent des adaptations similaires en réponse à des environnements équivalents (Schluter 2000). L'analyse des traits morphologiques a permis de constater que les différences morphologiques entre les écotypes semblent varier d'un lac à l'autre et que, dans l'ensemble, les différences morphologiques sont plus importantes entre les individus de lacs distincts qu'entre les écotypes d'un même lac. Ce résultat suggère un effet important des conditions environnementales locales de chaque lac sur l'expression phénotypique des ombles de fontaine (Sigursteinsdóttir et Kristjánsson 2005). Bien que les différences morphologiques entre les écotypes se sont avérées subtiles, les analyses de trajectoire de changement phénotypique (PCVA) entre les écotypes ont tout de même révélé un certain degré d'évolution parallèle où l'écotype littoral est plus trapu comparativement à l'écotype pélagique (15 populations sur 18). Ces faibles niveaux de parallélisme rappellent des résultats similaires où l'étendue du parallélisme entre les écotypes littoral et pélagique s'est avérée très variable entre les populations de la même

espèce (Oke et al., 2017). Ainsi, les faibles niveaux d'évolution parallèle observés dans notre système d'étude, entre les écotypes littoral et pélagique, pourraient être le résultat d'un stade précoce de diversification ainsi que du haut niveau de plasticité et de la faible intégration morphologique que présente l'omble de fontaine (Peres-Neto et Magnan 2004).

Les travaux menés dans le cadre de cette thèse auront certainement une incidence importante en écologie fonctionnelle et évolutive. En effet, l'approche à grande échelle (c.-à-d., l'étude de près d'une trentaine de populations) que nous avons mise de l'avant a permis de généraliser et d'améliorer la compréhension théorique des mécanismes responsables de l'expression du polymorphisme associé aux ressources chez une espèce affichant un polymorphisme associé aux ressources qui est subtil. En effet, l'omble de fontaine lacustre offre un système de recherche particulièrement intéressant pour la compréhension des processus de spéciation sympatrique, car ces populations semblent se situer au tout début du continuum de spéciation sympatrique (Hendry 2009) et donc représentent une opportunité d'explorer les tout premiers facteurs et mécanismes responsables de la diversification phénotypique menant au polymorphisme associé aux ressources et éventuellement à la spéciation.

5.2 Perspectives de recherche

Plusieurs questions sont demeurées sans réponse suite aux travaux de recherche menés dans le cadre de cette thèse et certains résultats ont même soulevé de nouveaux questionnements. Dans les limites de la présente démarche, nous nous sommes principalement attardés aux spécialisations phénotypiques entre les écotypes (c.-à-d., spécialisations morphologiques, alimentaires et dans la sélection d'habitats) ainsi qu'à leurs déterminants. Cependant, comme le souligne Hendry (2009), pour que le polymorphisme associé aux ressources puisse engendrer une spéciation, les spécialisations et différences dans l'écologie des écotypes doivent induire un isolement reproducteur entre les écotypes. Or, cet aspect a encore été très peu étudié dans ce système et a montré que le degré d'isolement reproducteur entre les écotypes littoral et pélagique est variable d'une population à l'autre, et ce malgré la présence de différences

morphologiques entre les écotypes dans les deux populations à l'étude (Dynes et al. 1999). En association avec nos résultats, il serait donc intéressant d'identifier les facteurs environnementaux et écologiques associés au degré d'isolement reproducteur entre les écotypes pour ainsi mieux comprendre quels sont les éléments clés permettant à la plasticité phénotypique (morphologique et comportementale) d'induire des différences génétiques entre écotypes. En effet, les avancées technologiques et méthodologiques récentes d'analyses génétiques ont, entre autres, permis des avancées majeures dans la compréhension de l'architecture génomique sous-jacente aux traits impliqués dans la divergence phénotypique (Skúlason et al. 2019). Étant à un stade de diversification très précoce, où le degré de différenciation morphologique (Bertrand et al. 2008; chapitre IV), comportementale et génétique (Dynes et al. 1999) entre les écotypes est variable d'une population à l'autre, l'omble de fontaine offre l'occasion d'explorer les facteurs limitant ou facilitant le processus de spéciation. En effet, la confirmation que la réduction du flux génétique entre les écotypes est le résultat d'adaptation à des habitats ou ressources différentes constituerait une avancée majeure en écologie évolutive (Hendry 2009).

Bien que nous ayons inféré l'effet de la compétition intraspécifique sur l'utilisation des ressources dans le chapitre II, nous n'avons pas été en mesure, pour des raisons logistiques, d'inclure dans notre plan d'échantillonnage des lacs où la pêche sportive est interdite ou absente. En effet, plusieurs études portant sur les causes de la diversification phénotypique et du polymorphisme associé aux ressources identifient les fortes pressions de compétition intraspécifique comme un déterminant fondamental (p. ex., Bolnick 2001; Huss et al. 2008; Svanbäck et al. 2009). Comme la pression de pêche tend à réduire la compétition intraspécifique des populations d'omble de fontaine (Magnan et al. 2005), il est possible que les forces de diversification soient maintenues à des niveaux plus faibles dans les lacs de la présente étude en raison de cette exploitation anthropique. Il serait donc intéressant d'explorer l'effet maximal de la compétition intraspécifique en comparant les résultats de la présente étude à des lacs où l'exploitation humaine est négligeable, voire absente.

5.3 Conclusion

Cette thèse a examiné comment le niveau de compétition interspécifique ainsi que plusieurs facteurs environnementaux affectent l'expression du polymorphisme associé aux ressources chez l'omble de fontaine lacustre du Bouclier laurentien. Nous y avons développé un cadre conceptuel basé sur les isodars qui nous a permis d'étudier les déterminants de la sélection d'habitats chez l'omble de fontaine. Nous y avons également explicitement testé l'hypothèse de l'opportunité écologique qui stipule que la variation phénotypique devrait être plus élevée suite au relâchement de la compétition interspécifique. Pour ce faire, cette thèse s'est appuyée sur un échantillonnage extensif de près d'une trentaine de populations d'omble de fontaine lacustre et sur des analyses statistiques tenant compte de la structure hiérarchique des données. Les contributions principales de cette thèse sont 1) d'avoir confirmé l'effet de la compétition intraspécifique sur l'utilisation des habitats littoral et pélagique par l'omble de fontaine, 2) d'avoir généralisé que la spécialisation alimentaire entre les écotypes littoral et pélagique est un phénomène commun dans les lacs du Bouclier laurentien et que contrairement à ce que l'hypothèse de l'opportunité écologique suggère, le niveau de spécialisation alimentaire des deux écotypes est demeuré constant en réponse au relâchement de la compétition interspécifique, 3) d'avoir observé un certain niveau d'évolution parallèle entre les écotypes de plusieurs populations où les individus littoraux sont caractérisés par un corps en moyenne plus trapu que les individus pélagiques. Ainsi, cette thèse apporte non seulement une meilleure compréhension des déterminants de l'expression du polymorphisme associé aux ressources, mais a également permis de généraliser les connaissances acquises dans ce système qui étaient, jusqu'ici limitées à un nombre restreint de lacs.

RÉFÉRENCES BIBLIOGRAPHIQUES

Araújo MS, Bolnick DI, Layman CA (2011) The ecological causes of individual specialisation. Ecology Letters 14: 948-958. doi: 10.1111/j.1461-0248.2011.01662.x

Barluenga M, Stölting KN, Salzburger W, Muschick M, Meyer A (2006) Sympatric speciation in nicaraguan crater lake cichlid fish. Nature. 439: 719-723.

Bertolo A, Pépino M, Adams J, Magnan P (2011) Behavioural thermoregulatory tactics in lacustrine brook charr, *Salvelinus fontinalis*. PLoS One 6:e18603. doi: 10.1371/journal.pone.0018603

Bertrand M, Marcogliese DJ, Magnan P (2008) Trophic polymorphism in brook charr revealed by diet, parasites and morphometrics. Journal of Fish Biology 72: 555-572. doi: 10.1111/j.1095-8649.2007.01720.x

Bolnick DI (2001) Intraspecific competition favours niche width expansion in *Drosophila melanogaster*. Nature 410: 463-466.

Bolnick DI, Svanbäck R, Fordyce JA, Yang LH, Davis JM, Hulsey CD, Forister, ML (2003) The ecology of individuals: incidence and implications of individual specialization. The American Naturalist 161: 1-28. doi: 10.2307/3078879

Bolnick DI, Ingram T, Stutz WE, Snowberg LK, Lau OL, Paull JS (2010) Ecological release from interspecific competition leads to decoupled changes in population and individual niche width. Proceedings of the Royal Society B 277: 1789-1797. doi: 10.1098/rspb.2010.0018

Bourke P, Magnan P, Rodriguez MA (1997) Individual variations in habitat use and morphology in brook charr. Journal of Fish Biology 51: 783-794.

Bourke P, Magnan P, Rodriguez MA (1999) Phenotypic responses of lacustrine brook charr in relation to the intensity of interspecific competition. Evolutionary Ecology 13: 19-31.

Dall SRX, Bell AM, Bolnick DI, Ratnieks FLW (2012) An evolutionary ecology of individual differences. Ecology Letters 15: 1189-1198. doi: 10.1111/j.1461-0248.2012.01846.x

Darwin C (1859) On the origin of species by means of natural selection. London: John Murray.

Dynes J, Magnan P, Bernatchez L, Rodriguez MA (1999) Genetic and morphological variation between two forms of lacustrine brook charr. Journal of Fish Biology 54: 955-972.

Ehlinger TJ (1990) Habitat choice and phenotype-limited feeding efficiency in bluegill: individual differences and trophic polymorphism. Ecology 71: 886-896. doi: 10.2307/1937360

Goyer K, Bertolo A, Pépino M, Magnan P (2014) Effects of lake warming on behavioural thermoregulatory tactics in a cold-water stenothermic fish. PLoS One 9:e92514. doi: 10.1371/journal.pone.0092514

Halliday WD, Blouin-Demers G (2017) Can temperature modify the strength of density-dependent habitat selection in ectotherms? A test with red flour beetles. Journal of Zoology 304: 159-168. doi: 10.1111/jzo.12510

Hendry AP (2009) Ecological speciation! Or the lack thereof? Canadian Journal of Fisheries and Aquatic Sciences 66: 1383-1398. doi: 10.1139/F09-074

Hendry AP (2016) Eco-evolutionary Dynamics. Princeton University Press, Princeton, NJ.

Hindar K, Jonsson B (1982) Habitat and food segregation of dwarf and normal arctic charr (*salvelinus alpinus*) from Vangsvatnet lake, western Norway. Canadian Journal of Fisheries and Aquatic Sciences. 39: 1030-1045. doi: 10.1139/f82-138

Huss M, Byström P, Persson L (2008) Resource heterogeneity, diet shifts and intra-cohort competition: effects on size divergence in YOY fish. Oecologia 158: 249-257. doi: 10.1007/s00442-008-1140-9

Hutchinson GE (1957) Concluding remarks. Cold Spring Harbor Symposia on Quantitative Biology 22: 415-427. doi: :10.1101/SQB.1957.022.01.039

Knudsen R, Primicerio R, Amundsen P-A, Klemetsen A (2010) Temporal stability of individual feeding specialization may promote speciation. Journal of Animal Ecology 79: 161-68. doi: 10.1111/j.1365-2656.2009.01625.x

MacArthur RH, Diamond JM, Karr JR (1972) Density Compensation in Island Faunas. Ecology53: 330-342.

Magnan P (1988) Interaction between Brook Charr, *Salvelinus fontinalis*, and nonsalmonid species: Ecological shift, morphological shift, and their impact on zooplankton communities. Canadian Journal of Fisheries and Aquatic Sciences 45: 999-1009.

Magnan P, Proulx R, Plante M (2005) Integrating the effects of fish exploitation and interspecific competition into current life history theories: an example with lacustrine brook trout (Salvelinus fontinalis) populations. Canadian Journal of Fisheries and Aquatic Sciences 62: 747-757. doi: 10.1139/F05-041.

Malmquist HJ, Snorrason SS, Skulason S, Jonsson B, Sandlund OT, Jonasson PM (1992) Diet differentiation in polymorphic arctic charr in Thingvallavatn, Iceland. Journal of Animal Ecology 61: 21-35.

Marchand F, Magnan P, Boisclair D (2002) Water temperature, light intensity and zooplankton density and the feeding activity of juvenile brook charr (*Salvelinus fontinalis*). Freshwater ecosystem. 47: 2153-62. doi: 10.1046/j.1365-2427.2002.00961.x

Martin RA, Pfennig DW (2010) Field and experimental evidence that competition and ecological opportunity promote resource polymorphism. Biological Journal of the Linnean Society 100: 73-88. doi: 10.1111/j.1095-8312.2010.01380.x

Morris DW (1988) Habitat-dependent population regulation and community structure. Evolutionary Ecology 2: 253-269.

Nakazawa T (2015) Ontogenetic niche shifts matter in community ecology: a review and future perspectives. Population Ecology 57 : 347-354. doi: 10.1007/s10144-014-0448-z

Nosil P, Reimchen TE (2005) Ecological opportunity and levels of morphological variance within freshwater stickleback populations. Biological Journal of the Linnean Society 86: 297-308. doi: 10.1554/04-428

Oke KB, Rolshausen G, LeBlond C, Hendry AP (2017) How parallel is parallel evolution? A comparative analysis in fishes. The American Naturalist 190: 1-16 doi: 10.1086/691989

O'Reilly CM, Sharma S, Gray DK, Hampton SE, Read JS, Rowley RJ et al. (2015) Rapid and highly variable warming of lake surface waters around the globe. Geophysical Research Letters 42: 10773-10781. Doi: 10.1002/2015GL066235

Peres-Neto PR, Magnan P (2004) The influence of swimming demand on phenotypic plasticity and morphological integration: a comparison of two polymorphic charr species. Oecologia 140: 36-45. doi: 10.1007/s00442-004-1562-y

Pfennig DW (1992) Polymorphism in spadefoot toad tadpoles as a logically adjusted evolutionarily stable strategy. Evolution 46: 1408-1420. doi: 10.2307/2409946

Pfennig DW, Wund MA, Snell-Rood EC, Cruickshank T, Schlichting CD, Moczek AP (2010) Phenotypic plasticity's impacts on diversification and speciation. Trends in Ecology and Evolution 25: 459-467. doi: 10.1016/j.tree.2010.05.006

Proulx R, Magnan P (2002) Physiological performance of two forms of lacustrine brook charr, *Salvelinus fontinalis*, in the open-water habitat. Environmental Biology of Fishes 64: 127-136.

Proulx R, Magnan P (2004) Contribution of phenotypic plasticity and heredity to the trophic polymorphism of lacustrine brook charr (*Salvelinus fontinalis* M.). Evolutionary Ecology Research 6: 503-522.

Robinson BW, Wilson DS (1994) Character release and displacement in fishes: a neglected literature. Am Nat 144: 596-627.

Roughgarden J (1972) Evolution of niche width. The American Naturalist 106: 683-718.

Roughgarden J (1974) Niche Width: Biogeographic Patterns Among Anolis Lizard Populations. The American Naturalist 108: 429-442.

Roughgarden J (1979) Theory of population genetics and evolutionary ecology: An introduction. Macmillan, New York, USA.

Rouleau S, Glémet H, Magnan P (2010) Effects of morphology on swimming performance in wild and laboratory crosses of brook trout ecotypes. Functional Ecology 24: 310-321. doi: 10.1111/j.1365-2435.2009.01636.x

Sacotte S, Magnan P (2006) Inherited differences in foraging behaviour in the offspring of two forms of lacustrine brook charr. Evolutionary Ecology Research 8: 843-857.

Schluter D (1995) Adaptive radiation in sticklebacks: trade-offs in feeding performance and growth. Ecology. 76: 82-90. doi: 10.2307/1940633

Schluter D (2000) The ecology of adaptive radiation. Oxford Univ. Press, Oxford.

Schluter D, McPhail JD (1993) Character displacement and replicate adaptive radiation. Trends in Ecology & Evolution 8: 197-200. doi: 10.1016/0169-5347(93)90098-A

Shine R (1989) Ecological causes for the evolution of sexual dimorphism: A review of the evidence. Quarterly Review of Biology 64:419-461.

Sigursteinsdóttir RJ, Kristjánsson BK (2005) Parallel evolution, not always so parallel: Comparison of small benthic charr, Salvelinus alpinus, from Grímsnes and Thingvallavatn, Iceland. Environmental Biology of Fishes 74: 239-244. doi: 10.1007/s10641-005-0499-2

Skúlason S, Parsons KJ, Svanbäck R, Räsänen K, Ferguson MM, Adams CE, Amundsen PA, Bartels P, Bean CW, Boughman JW, Englund G, Guðbrandsson J, Hooker OE, Hudson AG, Kahilainen KK, Knudsen R, Kristjánsson BK, Leblanc CAL, Jónsson Z, Öhlund G, Smith C, Snorrason SS (2019) A way forward with eco evo devo: an extended theory of resource polymorphism with postglacial fishes as model systems. Biological Reviews 94:1786-1808. doi: 10.1111/brv.12534

Skúlason S, Smith TB (1995) Resource polymorphisms in vertebrates. Trends in Ecology and Evolution 10:366-370.

Smith TB (1990) Comparative breeding biology of the two bill morphs of the black-bellied seedcracker (*Pyrenestes ostrinus*). The Auk. 107: 153-160.

Smith TB, Skúlason S (1996) Evolutionary significance of resource polymorphisms in fishes, amphibians, and birds. Annual Review of Ecology and Systematics 27: 111-133.

Snorrason SS, Skúlason S, Jonsson B, Malmquist HJ, Jonasson PM, Malmquist HJ, Jónasson PM, Sandlund OT, Lindem T (1994) Trophic specialization in arctic charr *Salvelinus alpinus* (pisces; salmonidae): morphological divergence and ontogenetic niche shifts. Biological Journal of the Linnean Society 52: 1-18. doi: 10.1006/bijl.1994.1035

Svanbäck R, Bolnick DI (2007) Intraspecific competition drives increased resource use diversity within a natural population. Proc Biol Sci 274:839-844. doi: 10.1098/rspb.2006.0198

Svanbäck R, Eklöv P, Fransson R, Holmgren K (2008) Intraspecific competition drives multiple species resource polymorphism in fish communities. Oikos 117: 114-124. doi: 10.1111/j.2007.0030-1299.16267.x

Svanbäck R, Quevedo M, Olsson J, Eklöv P (2015) Individuals in food webs: the relationships between trophic position , omnivory and among−individual diet variation. Oecologia 178: 103-114. doi: 10.1007/s00442-014-3203-4

Van Valen L (1965) Morphological Variation and Width of Ecological Niche. American Naturalist 99: 377-390.

Webb PW (1984) Body form, locomotion and foraging in aquatic vertebrates. American Zoologist 24: 107-20. doi: 10.1093/icb/24.1.107

Wellborn GA, Langerhans RB (2015) Ecological opportunity and the adaptive diversification of lineages. Ecology and Evolution 5: 176-195. doi: 10.1002/ece3.134. 7

Wilson DS (1998) Adaptive individual differences within single populations. Proceedings of the Royal Society B 353: 199-205.

Wilson ADM, McLaughlin RL (2007) Behavioural syndromes in brook charr, *Salvelinus fontinalis*: prey-search in the field corresponds with space use in novel laboratory situations. Animal Behaviour 74: 689-698. doi: 10.1016/j.anbehav.2007.01.009

Wilson AJ, Gislason D, Skúlason S, Snorrason S, Adams CE, Alexander G, Danzmann RG, Ferguson MM (2004) Population genetic structure of Arctic charr, Salvelinus alpinus from northwest Europe on large and small spatial scales. Molecular Ecology 13: 1129-1142. doi: 10.1111/j.1365-294X.2004.02149.x

Woolway RL, Merchant CJ (2019) Worldwide alteration of lake mixing regimes in response to climate change. Nature Geoscience 12: 271-276. Doi: 10.1038/s41561-019-0322-x

Yoder, JB, Clancey E, Des Roches S, Eastman JM, Gentry L, Godsoe W, Hagey TJ, Jochimsen D, Oswald BP, Robertson J, Sarver BAJ, Schenk JJ, Spear SF, Harmon LJ (2010) Ecological opportunity and the origin of adaptive radiations. Journal of Evolutionary Biology 23: 1581-1596. doi: 10.1111/j.1420-9101.2010.02029.x